J Comp Physiol A (1997) 181: 607–614

ORIGINAL PAPER

Y. Hosokawa · J. Horikawa · M. Nasu · I. Taniguchi

Real-time imaging of neural activity during binaural interaction in the guinea pig auditory cortex

Accepted: 14 August 1997

Abstract Spatio-temporal patterns of binaural interaction in the guinea pig auditory cortex (AC) were observed using optical recording with a 12×12 photodiode array and a voltage-sensitive dye. The amplitudes of the sound-induced light signals from the cortex were transformed into sequential two-dimensional images every 0.58 ms. Binaural sound stimuli evoked an excitatory response followed by a strong inhibition, and contralateral stimuli evoked a strong excitatory response followed by a weak inhibition. Ipsilateral sound stimuli evoked a weak response. Binaural stimulation induced two types of ipsilateral inhibition: a fast binaural inhibition which was detected only after the contralateral and ipsilateral responses were subtracted from the binaural responses, and which appeared 12–25 ms after the onset of stimulation, and a slow binaural inhibitory effect which was clearly observed in the binaural responses themselves, appearing 70–95 ms after the onset of stimulation. The fast binaural inhibition was observed in the same area as the contralateral excitatory response. The inhibited area became stronger and more widespread with increasing intensity of ipsilateral stimulation. We did not observe the specialized organization of binaural neurons as electrophysiologically found in the cat AC, in which binaural neurons of the same binaural response type are clustered together and alternate with clusters of other response types.

Key words Binaural responses · Optical recording · Guinea pig · Auditory cortex

Abbreviations *A* anterior · *AC* auditory cortex · *AI* primary auditory cortex · *DC* dorso-caudal · *ECG* electrocardiograph · *EE* excitation of each ear · *EE/F* binaural facilitation · *EE/I* binaural inhibition · *EI* contralateral excitation and ipsilateral inhibition type · *EPSP* excitatory postsynaptic potential · *GABA* γ-aminobutyric acid · *IID* interaural intensity difference · *SPL* sound pressure level

Y. Hosokawa (✉) · J. Horikawa · M. Nasu · I. Taniguchi
Department of Neurophysiology, Medical Research Institute, Tokyo Medical and Dental University,
2-3-10 Kanda-surugadai, Chiyoda-ku, Tokyo, 101, Japan
Fax: +81-3-5280-8073
e-mail: hosokawa.nphy@mri.tmd.ac.jp

Introduction

Many electrophysiological studies on binaural interactions in the auditory cortex (AC) have been performed in various species, such as cats (Imig and Adrian 1977; Phillips and Irvine 1979, 1983; Middlebrooks et al. 1980; Schreiner and Cynader 1984; Reale and Kettner 1986; Imig et al. 1990; Rajan et al. 1990; Samson et al. 1993, 1994; Semple and Kitzes 1993a,b; Irvine et al. 1996), ferrets (Kelly and Judge 1994), rats (Kelly and Sally 1988), chinchillas (Benson and Teas 1976), bats (Manabe et al. 1978), macaque monkeys (Brugge and Merzenich 1973), and gerbils (Heffner and Heffner 1988; Caird et al. 1991).

Historically, binaural neurons have been classified into two major response types: neurons excited by each-ear stimulation (EE type) and those excited by contralateral stimulation and inhibited by ipsilateral stimulation (EI type). The EE neurons were reported to be segregated from the EI neurons, occurring in alternating bands along the isofrequency bands (Imig and Adrian 1977; Middlebrooks et al. 1980; Schreiner and Cynader 1984). However, Phillips and Irvine (1979, 1983) argued that the EE and EI response-type dichotomy involved a considerable oversimplification of the complexities of the binaural input and obscured the diversity of the binaural response types.

Recent studies have reported a more detailed classification based on inhibitory, facilitatory, and mixed facilitatory-inhibitory binaural interactions, and have shown that the different types of binaural neurons are indeed segregated but form patches rather than alternating bands (rats: Kelly and Sally 1988; cats: Reale and Kettner 1986). Nevertheless, Kelly and Judge (1994)

have reported the existence of an alternating band structure between binaural facilitation (EE/F) and inhibition (EO/I) neurons along an extension of the isofrequency contours, with mixed-type neurons located only at the border between them.

It has been reported that the relative intensities of binaural facilitation and inhibition are dependent upon the average binaural intensity and the interaural intensity differences (IIDs) which provide an important cue to sound localization in mammals (Imig et al. 1990; Rajan et al. 1990; Samson et al. 1993, 1994; Semple and Kitzes 1993a,b), and that the azimuthal sensitivity corresponds closely with IID sensitivity (Irvine et al. 1996). Studies of sound localization in the primary auditory cortex (AI) neurons of cats have also shown that neurons with different types of azimuthal sensitivity display a complex but non-random distribution pattern along the isofrequency bands of AI (Rajan et al. 1990; Clarey et al. 1994). Relatively little is known about the binaural organization of the AC in other mammals such as mustached bats, macaque monkeys, chinchillas, and gerbils, although segregation of a binaural response area has been reported (Brugge and Merzenich 1973; Benson and Teas 1976; Mahabe et al. 1978; Heffner and Heffner 1988; Caird et al. 1991).

Recently the spatio-temporal representation of sound frequency and intensity in the AC of the guinea pig has been observed by optical recording (Taniguchi et al. 1992, 1993). Using this method, we have performed real-time imaging of neuronal activity during binaural interactions in the guinea pig AC. We have investigated whether neurons of different binaural types are segregated, and how the binaural responses change with different IIDs. Such an approach provides another view into the functional organization of the AC with regard to the dynamics of neuronal activation in binaural hearing.

Materials and methods

Animal preparation and recording procedures have been described in detail elsewhere (Taniguchi et al. 1992), and will be outlined only briefly here.

Animal preparation

Fifteen guinea pigs (280–450 g) were anesthetized intraperitoneally with sodium pentobarbital (30 mg kg^{-1}). Anesthesia was maintained with supplemental doses of neuroleptic solutions: droperidol (16.7 mg $kg^{-1}h^{-1}$) and pentazocine (5 mg kg h^{-1}). The external auditory meatus was exposed following cannulation of the trachea and placing an electrode onto the animal's breast for monitoring the electrocardiogram (ECG). The guinea pig's head was clamped in a stereotaxic frame using hollow earbars. The AC was exposed via a small hole drilled in the temporal bone. The dura and arachnoid membranes were removed. The exposed AC was covered with a voltage-sensitive dye, RH795 (0.2 mg ml^{-1} diluted in saline; Molecular Probes, USA; Grinvald et al. 1986) for 90 min. The experiments were performed under artificial ventilation after intraperitoneal injection of a muscle relaxant (pancuronium bromide, 1 mg kg^{-1} h^{-1}) in addition to droperidol and pentazocine. Pure O_2 gas was added to the air in the respirator. The experiments were carried out in a sound-proof room.

Stimulation

Pure tones (100 ms duration, 10-ms rise/fall time) and white noise (0.1–25 kHz bandwidth, 100 ms duration, 10-ms rise/fall time) were used as acoustic stimuli. The tones were generated by two multifunction synthesizers (1930A, NF Electronic Instruments, Japan) and shaped by laboratory-made electronic switches. The noise stimuli were generated by a sound synthesizer system (Signal-RTS, Engineering Design, USA). The sound signals were triggered by the ECG and fed to condenser earphones (Murata et al. 1986) through power amplifiers (2713, Brüel & Kjaer, Denmark) and attenuators (TPA-114A, Tama Electronic Instruments, Japan). The sound stimuli were delivered at a rate of about 1 s^{-1}. The intensity of stimuli delivered to each ear could be controlled independently. The frequency characteristics of the earphones fixed at both ears were measured automatically using a personal computer (9801RA, NEC, Japan) at 360 points between 0.1 kHz and 99 kHz, using a calibrated probe tube microphone (4134, Brüel & Kjaer, Denmark) and a level meter (2619 and 2636, Brüel & Kjaer, Denmark). The frequency characteristics were stored in the personal computer. The sound pressure was expressed as dB sound pressure level (SPL) re. 20 μPa. Noise intensities were expressed as peak equivalent pressures, i.e., the intensity (dB SPL) required for a 1-kHz pure tone to match the peak-to-peak amplitude of the acoustic waveform of the noise.

To test the dependency of the binaural response in the AC on the sound intensity present in the ear ipsilateral to the cortex, we fixed the contralateral intensity and changed the ipsilateral one. The IID was expressed as the value of the contralateral intensity with reference to the ipsilateral intensity so that negative values of IID imply that the ipsilateral intensity was greater than the contralateral intensity.

Acoustic crosstalk between the left and right ears was measured in two animals by examining cochlear microphonic potentials during ipsilateral and contralateral stimulation. These measurements indicated that the interaural separation was at least 50 dB for frequencies between 100 Hz and 25 kHz.

Recording

An epifluorescence microscope (LW-101, Hiland, Japan) with a 150-W tungsten-halogen lamp was used for optical recording. Transmitted fluorescence at 620 nm from the cortex was detected by a 12 × 12 photodiode array. The optical signals $\Delta F/F$ (F: light intensity at rest, ΔF: change in intensity induced by neural signals) were sampled using an A/D converter (16-bit, 4-μs sampling rate; Narumi Electric, Japan). Two types of trial, with and without stimulus, were triggered by the peak of the ECG. The difference between the two trials, canceling out the pulsation noise, was calculated on a personal computer (386GS, Epson, Japan). The optical signals were averaged over ten trials. To observe the spatio-temporal pattern of neural activity, the response amplitudes at 144 locations were color coded and transformed into sequential two-dimensional images every 0.58 ms. The images were smoothed, both temporally and spatially.

The general organization of the guinea pig AC into two fields, the anterior (A) and dorsocaudal (DC) fields, and the positions of the border between the fields were determined in each animal by presentation of low-intensity tone bursts (contralateral stimulation) as described before (Taniguchi et al. 1992). The mean latencies of sound responses in fields A and DC were calculated from some optical signals in each field.

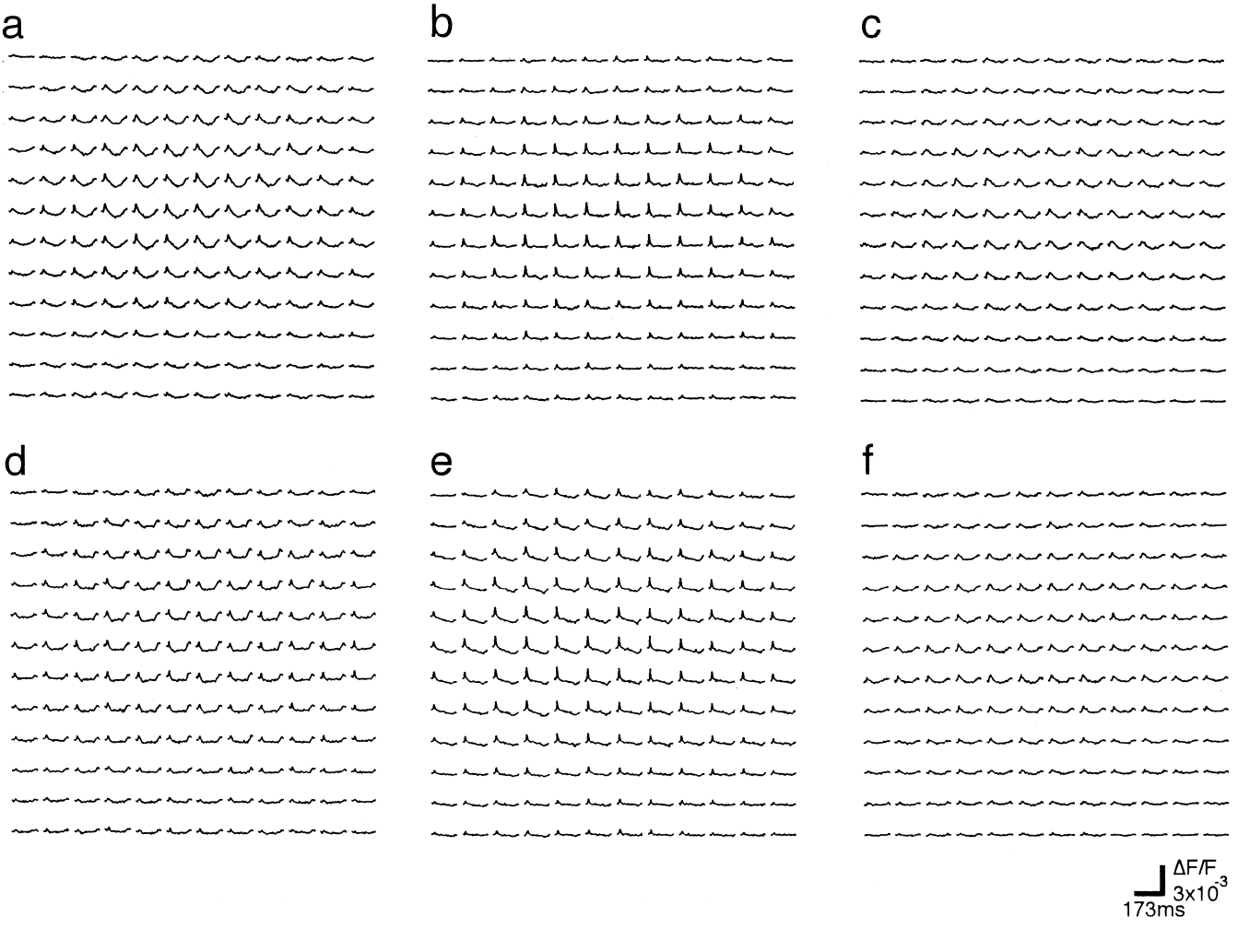
Results

Optical signals in response to binaural and monaural stimulation

Representative optical signals from the 144 photodiode elements to binaural and monaural tone-burst stimulation (14 kHz, 75 dB SPL) are shown in Fig. 1a,b,c. When the ears were binaurally stimulated at the same intensity, strong responses were localized in an area that depended on the sound frequency. The binaural response consisted of a weak excitatory component followed by a strong inhibitory component (Fig. 1a). On stimulation of the contralateral ear alone, a strong excitatory component followed by a weak inhibitory component was observed. The amplitude of the contralateral excitatory component was larger than that of the binaural one, and the amplitude of the contralateral inhibitory component was smaller than that of the binaural one (Fig. 1b). On stimulation of the ipsilateral ear alone, a weak excitatory component was followed by a weak inhibitory component (Fig. 1c). The latency of the ipsilateral excitatory component (15.7 ± 0.9 ms; $n = 8$ channels) was longer than those of the contralateral and the binaural excitatory components (13.8 ± 0.4 ms; $n = 8$).

The optical signals produced in response to white-noise stimulation (0.1–25 kHz) are shown in Fig. 1d,e,f. When the ears were stimulated binaurally, the optical signals appeared over a wider area along the rostro-caudal axis than those to tone-burst stimulation (Fig. 1d). The binaural responses to noise stimulation consisted of a weak excitatory component followed by a strong inhibitory component, similar to the binaural response to tone-burst stimulation. The latency of the inhibitory component of the binaural noise response (32.8 ± 1.3 ms; $n = 8$) was shorter than that of the

Fig. 1a–f Optical signals in response to monaural and binaural sound stimuli: **a** binaural tone-burst stimulation [14 kHz, 75 dB SPL, IID: 0 dB (dB; contra. re ipsi.)]; **b** contralateral tone-burst stimulation only (14 kHz, 75 dB SPL); **c** ipsilateral tone-burst stimulation only (14 kHz, 75 dB SPL); **d** binaural noise stimulation [0.1–25 kHz, 85 dB peak equivalent (p. e.), IID: 0 dB]; **e** contralateral noise stimulation only (0.1–25 kHz, 85 dB p. e.); **f** ipsilateral noise stimulation only (0.1–25 kHz, 85 dB p. e.). Note that two kinds of inhibitory effects are observed



binaural tone-burst response (61.0 ± 2.8 ms; $n = 8$). On stimulation of the contralateral ear alone by noise, a sharp and strong excitatory component was followed by a weak inhibitory component (Fig. 1e). The time-course of the inhibitory component of the contralateral noise response was steeper than that of the binaural noise response. On stimulation of the ipsilateral ear by noise alone, a weak excitatory component was followed by a inhibitory component (Fig. 1f). The waveform of the ipsilateral noise response was similar to that of the ipsilateral tone-burst response.

To reveal the inhibitory effects induced by binaural stimulation, the contralateral and ipsilateral responses were subtracted from the binaural response. A typical example is shown in Fig. 2. In most of the cases the waveform of the subtracted traces was composed of two troughs (Fig. 2d). The first and second troughs began with a latency of 19.3 ± 5.5 ms ($n = 14$ guinea pigs) and 89.1 ± 15.2 ms ($n = 14$), respectively, in field A. Their respective values in field DC was 20.3 ± 7.3 ms ($n = 14$) and 86.6 ± 15.0 ms ($n = 14$). The first trough lasted about 30 ms and the second trough lasted about 80 ms. As shown in Fig. 2a,b,c, the first trough represents the inhibition (or occlusion) induced by binaural stimulation, because in this phase only the excitatory response to each-ear stimulation was observed and the excitatory response to binaural stimuli was smaller than the sum of the contralateral and ipsilateral excitatory responses. The second trough represents the enhancement of the inhibitory component by binaural stimuli because in this phase only the inhibitory response to each-ear stimulation was observed and the inhibitory response to binaural stimulation was larger than the sum of the contralateral and ipsilateral inhibitory responses. Hereafter we call these first and second components fast and slow binaural inhibitions, respectively.

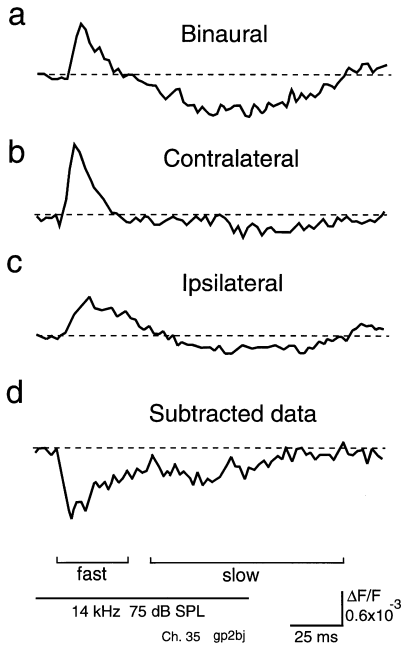


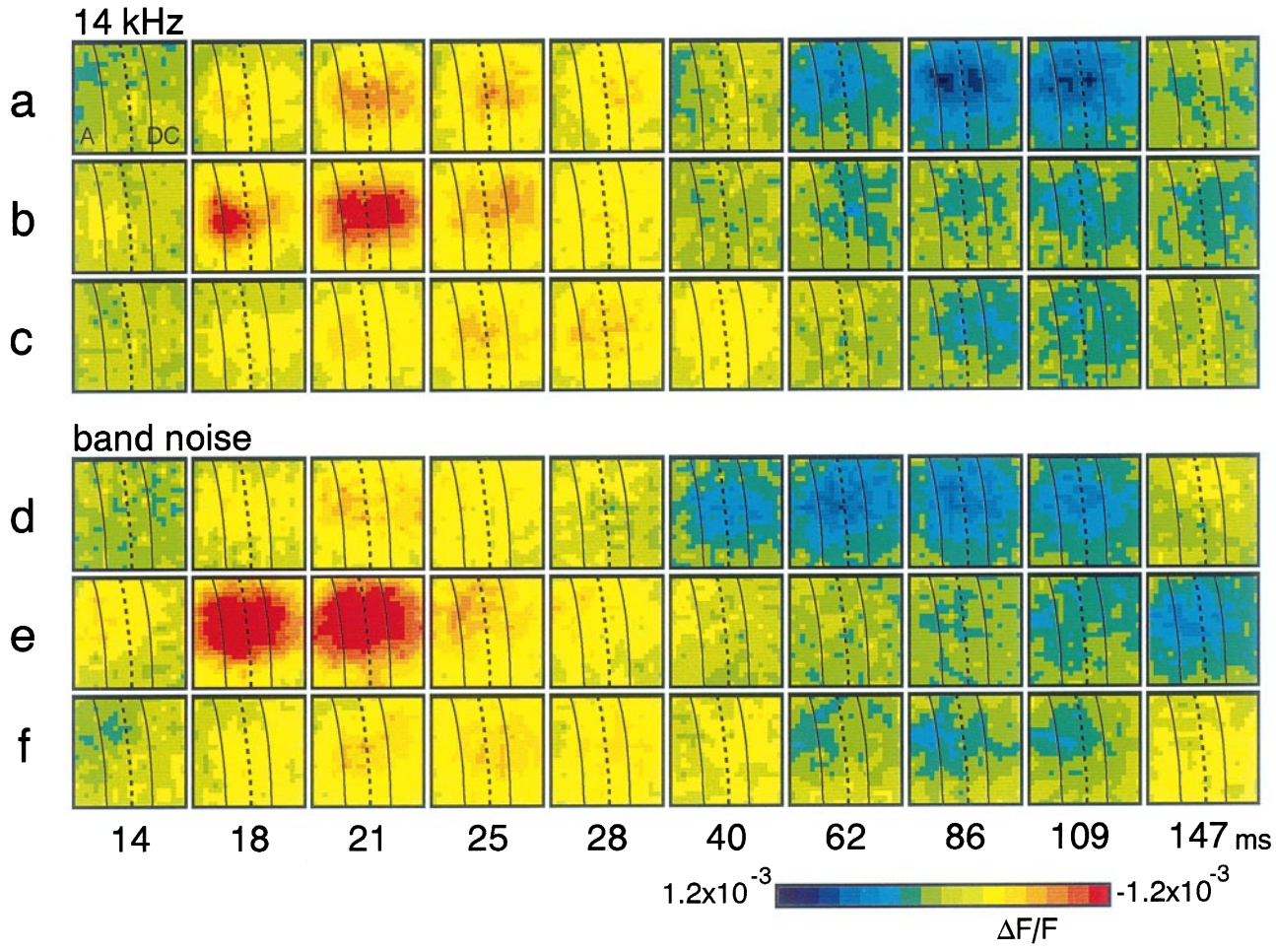
Fig. 2 a A typical optical signal evoked by the binaural tone stimuli picked up from Fig. 1a. IID value was 0 dB. **b** A typical optical signal evoked by the contralateral tone stimuli picked up from Fig. 1b. **c** A typical optical signal evoked by the ipsilateral tone stimuli picked up from Fig. 1c. **d** Subtraction of the contralateral and ipsilateral responses from the binaural one. *Fast* a fast binaural inhibition; *slow* a slow binaural inhibition. *Horizontal dashed lines*: resting level; *horizontal bar*: 100 ms long tone burst

Spatio-temporal images of responses to binaural and monaural stimulation

To obtain spatio-temporal images of the binaural and monaural responses, the amplitude of each optical signal from the 144 photodiode elements, seen in Fig. 1, was color coded and transformed into sequential two-dimensional images every 0.58 ms.

The spatio-temporal images in response to tone-burst stimuli (14 kHz) are shown in Fig. 3a,b,c. In the case of binaural tone-burst stimulation (Fig. 3a), a weakly-activated spot appeared in field A, 14 ms after the onset of the stimuli and at 16 ms in field DC. These activated spots expanded along the isofrequency band and then merged and spread widely over the boundary line. The activated spots disappeared at the dorsal portion of field DC. The inhibited area appeared at 62 ms in field A and spread from field A to field DC. The stronger inhibition appeared 86 ms after the onset of stimulation, both in field A and field DC near the isofrequency bands, and disappeared after 147 ms. In the case of contralateral tone-burst stimulation (Fig. 3b), the strongly activated spot appeared at the onset of the excitatory component at the position of the respective isofrequency bands. The contralateral active spot was stronger than that of the binaural one. The movement along isofrequency bands of the active spot of the contralateral response was similar to that of the binaural response. After the disappearance of the active spot, the weakly inhibited region, whose strength was lower than that of the binaural one, spread broadly. In the case of ipsilateral tone-burst stimulation (Fig. 3c), the active spot appeared later and lasted longer than that of the contralateral or the binaural stimulation. The movement of the active spot of the ipsilateral response was similar to that of the binaural one. The ipsilaterally inhibited area was similar to that of the contralateral one, but smaller than that of the binaural one (Fig. 3c). The binaural interaction was observed as a reduction of the contralateral excitatory component and an enhancement of the slow inhibition. The area of the ipsilateral inhibitory effects coincided with the isofrequency bands.

The spatio-temporal patterns to noise stimulation are shown in Fig. 3d,e,f. In the case of binaural noise stimulation (Fig. 3d), the active spots appeared to be more scattered than those of the binaural tone-burst response. The active spot moved to the dorsal portion of fields A and DC. After the disappearance of the active spot, a strongly inhibited region expanded in fields A and DC at 40 ms. The area inhibited by the binaural



noise appeared faster than that inhibited by the contralateral tone-burst stimulation. In the case of contralateral noise stimulation (Fig. 3e), a stronger, and spatially wider active spot appeared at the onset of the contralateral response and moved a little way to the dorsal portion and then disappeared. The inhibited area appeared later compared with that of the binaural noise response. In the case of ipsilateral noise stimulation (Fig. 3f), the weak active spot appeared later than that of the binaural noise response, and moved slowly from field A to field DC. After the disappearance of the active spot, the inhibited area appeared later than in the binaural noise response, and expanded to field A (Fig. 3f). The magnitude of inhibition induced by the binaural noise was about half (51.9 ± 9.7%; $n = 8$ channels) of that of the binaural one.

Fig. 3a–f The magnitudes of the optical signals shown in Fig. 1 were color coded and transformed into two-dimensional images for the period of 14–147 ms from the onset of the sound; **a–f** show the two-dimensional images corresponding to the data from Figs. 1a–f, respectively. The neural activity evoked by (**a**) binaural tone-burst stimulation is weaker than that evoked by (**b**) contralateral tone-burst stimulation. Ipsilateral tone-burst stimulation alone evoked weak excitatory neural activity followed by inhibition (**c**). The neural activity evoked by binaural noise stimulation is wider than that evoked by contralateral tone-burst stimulation (**d**). The neural activity evoked by contralateral noise stimulation alone is stronger than that evoked by binaural noise stimulation (**e**). Ipsilateral noise stimulation evoked weak excitatory neural activity followed by inhibition (**f**). In **a** and **d**, IID values were 0 dB. *Dashed lines* indicate the border between fields A and DC. *Straight lines* indicate the isofrequency contour lines of 14 kHz in fields A and DC

Spatial pattern of the binaural inhibitory effect of IID

To observe the spatial pattern of the binaural inhibitory effect of IID, the amplitudes of responses to the contralateral and ipsilateral stimulation were subtracted from those of binaural responses and transformed into gray-scaled images. The images based on IIDs of tone-burst stimuli (14 kHz) are shown in Fig. 4. The contralateral stimulus intensity was fixed at 70 dB SPL, and the ipsilateral stimulus intensity was changed from 60 dB SPL to 90 dB SPL in steps of 10 dB SPL. At an IID of +10 dB, the binaural inhibitory effect was slight (Fig. 4a). At an IID of 0 dB, patches of the binaural inhibitory effect appeared in the same region where the excitatory spot of the contralateral response had been observed (Fig. 4b). At an IID of −10 dB, the intensity of the binaural inhibitory effect in the patches increased (Fig. 4c). At an IID of −20 dB, patches of the binaural inhibitory

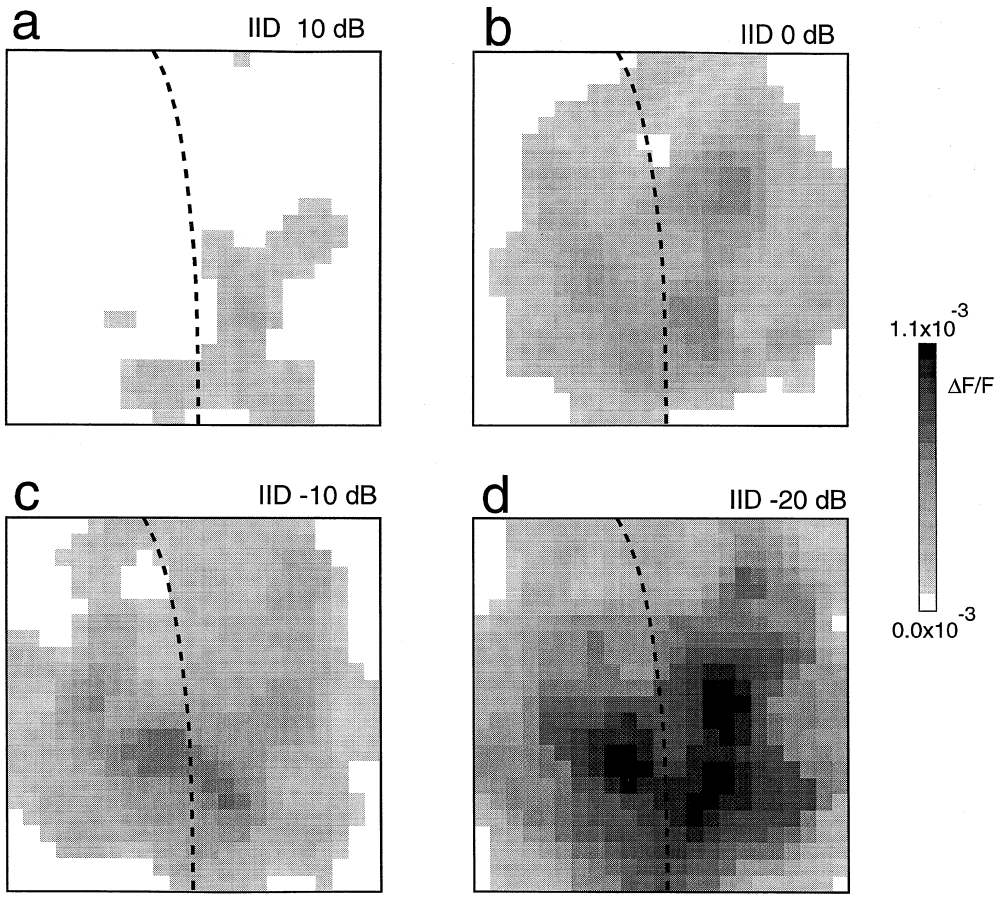


Fig. 4a–d Optical images of binaural inhibitory effects at 28 ms after sound stimulation to various IID levels. These data were obtained by subtracting the contralateral and ipsilateral responses from the binaural responses to tone bursts of 14 kHz. The response amplitudes were coded using a 16-level gray scale. The contralateral intensity was fixed at 70 dB SPL and the ipsilateral intensity was varied. **a** Images of the inhibitory effect of an IID of +10 dB **(a)**, 0 dB **(b)** −10 dB **(c)**, and −20 dB **(d)** (dB; contra re ipsi) are shown

effect expanded to the area surrounding the isofrequency band, and the inhibition magnitude increased (Fig. 4d). In other words, patches of the binaural inhibitory effect expanded, and their inhibition magnitude increased with decreasing IID. This tendency was observed also in the slow component of the binaural inhibitory effect (data not shown).

Discussion

Possible source of recorded optical signals

Previous reports have shown that the optical signals of neurons stained with voltage-sensitive dyes changed linearly with membrane potentials (Cohen and Lesher 1986; Grinvald et al. 1986; Salzberg and Obaid 1988). The optical signals observed in this study would be a result of the accumulated membrane potentials of many neurons because the recording cortical area was 250 μm × 250 μm and the effective focal depth of the microscope was about 200 μm. Individual spikes were not recorded since our sampling rate (about 0.6 ms) was not high enough. The optical signals to binaural stimulation observed in this study might include signals from different types of binaural neurons.

Subtraction of the contralateral and ipsilateral responses from the binaural responses were performed in order to observe the binaural interactions. The subtracted data indicate the interaction induced by binaural stimulation, as shown in Fig. 2.

Inhibitory effect in response to binaural stimulation

We observed both the fast and slow components of the inhibitory effects induced by binaural stimulation. The fast inhibitory component was observed as a reduction in the contralateral excitatory component. These results resemble the response patterns of EE/O neurons recorded using microelectrodes. That is, the neurons are excited by both monaural and binaural stimulation, and the binaural responses are smaller than the sum of contralateral and ipsilateral responses (chinchillas: Benson and Teas 1976; cats: Phillips and Irvine 1983; Reale and Kettner 1986; Irvine et al. 1996; rats: Kelly and Sally 1988). The fast component of the binaural inhibitory effect was an occlusion, and the inhibitory component was not observed in the responses to the ipsilateral stimulation alone.

A slow inhibitory component was observed in the monaural responses as well as in the binaural ones. Metherate and Ashe (1993, 1994), using in vivo whole-cell recording in the rat AC, reported that afferent stimulation of the medial geniculate body elicited a fast excitatory postsynaptic potential (EPSP) and a long-

lasting inhibitory postsynaptic potential (IPSP). The long-lasting IPSP consisted of polysynaptic early and late IPSPs. According to their results, the early IPSP mediated by gamma-aminobutyric acid $(GABA)_A$ receptors started at about 20 ms, and the late IPSP mediated by $GABA_B$ receptors started at about 120 ms. In our results, the slow inhibitory component started 20–30 ms after the onset of the excitatory component. This suggests that the slow inhibitory component observed in our study corresponds to the early IPSP reported by Metherate and Ashe, and that it is mediated by $GABA_A$ receptors. This is supported by the report that the inhibition seen in the guinea pig AC is mediated by $GABA_A$ receptors (Horikawa et al. 1996).

Spatial distribution of the inhibitory effect in response to binaural stimulation

Mapping studies of binaural interactions have been performed in several animals using microelectrode techniques. Such mapping experiments carried out in cats have revealed that neurons of the same binaural response type are clustered together, and that the clusters tend to extend rostrocaudally across the dorsoventrally oriented isofrequency contours in the AI (Imig and Adrian 1977; Middlebrooks et al. 1980; Schreiner and Cynader 1984). Recent studies of detailed classification in cats have reported that the different types of binaural neurons are segregated, but form patches and not the alternating band structure (Reale and Kettner 1986), although a study carried out using ferrets has shown an alternating structure between EE/F and EO/I neurons (Kelly and Judge 1994). In the AI of mustached bats (Manabe et al. 1978), rats (kelly and Sally 1988) and macaque monkeys (Brugge and Merzenich 1973), neurons of the same binaural response type aggregated but did not form the configuration of bands.

The study reported here has shown that the binaural inhibitory effect observed in guinea pigs occurs in the same regions as the contralateral excitatory responses do. Following tone-burst stimulation, the active spots of the contralateral response moved along the isofrequency bands and then spread into fields A and DC. The band structure found in the AI of cats and ferrets was not observed clearly in field A of guinea pigs. The area that was strongly inhibited by the slow binaural effect (compare Figs. 3a and 3d) was also observed near the isofrequency band where a contralateral sound evoked an excitatory response. During noise stimulation, the most strongly activated area was wider on the rostrocaudal axis than during the tone-burst stimulation, presumably because the noise contained many frequency components. The area of the fast and slow binaural inhibition caused by the noise appeared to be wider than that caused by the tone bursts. In these inhibited areas, we observed broad inhibition, but not the alternating structure that occurs in the cat and ferret.

In the cat, EE regions (200–3400 μm wide) and EI regions (100–2000 μm in width) changed across the AI (Middlebrooks et al. 1980). In the ferret, areas of EE/F or EO/I neurons were typically between 500 μm and 700 μm wide (Kelly and Judge 1994). If a similar binaural band structure exists in the guinea pig AC, and if its bandwidth is similar to that of cats and ferrets, it should have been observed by our optical recording device which has a spatial resolution of 250 μm × 250 μm. We did not see such alternating binaural response areas, therefore different types of binaurally responsive neurons may be distributed irregularly within the guinea pig AC, or arranged in small clusters or bands of less than 250 μm width.

IID dependency of the spatial pattern of the binaural inhibitory effect

We have found that decrements in IID cause an increase in the inhibitory response and an expansion in the inhibitory area. Reale and Kettner (1986), using electrophysiological methods, reported that the population of neurons in the AC of cats exhibiting suppression by binaural stimulation increases with decreasing IID. They also reported that the population of EO/I neurons increased and that of EE/F neurons decreased with decrements in IID. In most neurons of the mixed type, the binaural response changed from facilitatory to inhibitory type when the ipsilateral intensity was 10–30 dB louder than the contralateral one. The majority of binaural response types were inhibitory at IIDs smaller than −20 dB. Our work concurs with these results, showing that inhibition in the cortex becomes stronger, and the inhibitory area wider with increasing intensity of ipsilateral stimulation. Our data also agree with recent reports of the IID sensitivity of cortical neurons in the cat which show that the population of neurons sensitive to high IID values (contra-max type) is twice as large as that of neurons sensitive to low IID values (ipsi-max type), thus keeping the average binaural intensity constant (Irvine et al. 1996).

Recent studies of sound localization in AI neurons has shown that neurons with different types of azimuthal sensitivity are distributed non-randomly across the isofrequency direction of area AI (Clarey et al. 1994; Rajan et al. 1990). It has also been reported that the population of azimuthal-sensitive neurons is similar to that of IID-sensitive neurons (Irvine et al. 1996). These results implicate a topographical organization of IID sensitivity along the isofrequency band. However, our results did not show clear localization of the responses to IID, and showed only that the extent of the inhibitory area became larger with the decrements in IID value. These results suggest that in the guinea pig AC, neurons with different types of IID sensitivity are distributed irregularly or are arranged in small clusters.

Acknowledgements We thank Dr. G. Ehret for his critical comments on the early version of the manuscript. This work was supported by Grants-in-Aid for Scientific Research 06680784, 07680866, and 06454484 from the Ministry of Education, Science, Sports and Culture, Japan, a grant from the Nakatani Electronic Measuring Technology Association of Japan and the Human Frontier Science Program. These experiments comply with the guidelines of the Animal Experiments Committee of the Tokyo Medical and Dental University of Japan.

References

Benson DA, Teas DC (1976) Single unit study of binaural interaction in the auditory cortex of the chinchilla. Brain Res 103: 313–338

Brugge JF, Merzenich MM (1973) Responses of neurons in auditory cortex of the macaque monkey to monaural and binaural stimulation. J Neurophysiol 36: 1138–1158

Caird D, Scheich H, Klinke R (1991) Functional organization of auditory cortical fields in the mongolian gerbil (*Meriones unguiculatus*): binaural 2-deoxyglucose patterns. J Comp Physiol A 168: 13–26

Clarey JC, Barone P, Imig TJ (1994) Functional organization of sound direction and sound pressure level in primary auditory cortex of the cat. J Neurophysiol 72: 2383–2405

Cohen LB, Lesher S (1986) Optical monitoring of membrane potential: methods of multisite optical measurement. In: deWeer P, Salzberg MM (eds) Optical methods in cell physiology. Wiley, New York, pp 71–99

Grinvald A, Lieke E, Frostig RD, Gilbert C, Wiesel TN (1986) Functional architecture of cortex revealed by optical imaging of intrinsic signals. Nature 324: 361–364

Heffner RS, Heffner HE (1988) Sound localization and use of binaural cues by the gerbil (*Meriones unguiculatus*). Behav Neurosci 102: 422–428

Horikawa J, Hosokawa Y, Kubota M, Nasu M, Taniguchi I (1996) Optical imaging of spatiotemporal patterns of glutamatergic excitation and GABAergic inhibition in the guinea-pig auditory cortex in vivo. J Physiol (Lond) 497: 629–638

Imig TJ, Adrian HO (1977) Binaural columns in the primary field (AI) of cat auditory cortex. Brain Res 138: 241–257

Imig TJ, Irons WA, Samson FR (1990) Single-unit selectivity to azimuthal direction and sound pressure level of noise bursts in cat high-frequency primary auditory cortex. J Neurophysiol 63: 1448–1466

Irvine DRF, Rajan R, Aitkin LM (1996) Sensitivity to interaural intensity differences of neurons in primary auditory cortex of the cat. I. Types of sensitivity and effects of variations in sound pressure level. J Neurophysiol 75: 75–96

Kelly JB, Judge AP (1994) Binaural organization of primary auditory cortex in the ferret (*Mustela nutorius*). J Neurophysiol 71: 904–913

Kelly JB, Sally SL (1988) Organization of auditory cortex in the albino rat: binaural response properties. J Neurophysiol 59: 1756–1769

Manabe T, Suga N, Ostwald J (1978) Aural representation in the doppler-shifted-CF processing area of the auditory cortex of the mustache bat. Science 200: 339–342

Metherate R, Ashe JH (1993) Ionic flux contributions to neocortical slow waves and nucleus basalis-mediated activation: whole-cell recordings in vivo. J Neurosci 13: 5312–5323

Metherate R, Ashe JH (1994) Facilitation of an NMDA receptor-mediated EPSP by paired-pulse stimulation in rat neocortex via depression of GABAergic IPSPs. J Physiol (Lond) 481: 331–348

Middlebrooks JC, Dykes RW, Merzenich MM (1980) Binaural response-specific bands in primary auditory cortex (AI) of the cat: topographical organization orthogonal to isofrequency contours. Brain Res 181: 31–48

Murata K, Ito S, Horikawa J, Minami S (1986) The acoustic middle ear muscle reflex in albino rats. Hearing Res 23: 169–183

Phillips DP, Irvine DRF (1979) Methodological considerations in mapping auditory cortex: binaural columns in AI of cat. Brain Res 161: 342–346

Phillips DP, Irvine DRF (1983) Some features of binaural input to single neurons in physiologically defined area AI of cat cerebral cortex. J Neurophysiol 49: 383–395

Rajan R, Aitkin LM, Irvine DRF, McKay J (1990) Azimuthal sensitivity of neurons in primary auditory cortex of cats. I. Types of sensitivity and the effects of variations in stimulus parameters. J. Neurophysiol 64: 872–887

Reale RA, Kettner RE (1986) Topography of binaural organization in primary auditory cortex of the cat: effects of changing interaural intensity. J Neurophysiol 56: 663–682

Salzberg BM, Obaid AL (1988) Optical measurement of electrical and secretory events at vertebrate nerve terminals. In: Loew LM (ed) Spectroscopic membrane probes, vol III. CRC Press, Florida, pp 67–99

Samson FK, Clarey JC, Barone P, Imig TJ (1993) Effects of ear plugging on single-unit azimuth sensitivity in cat primary auditory cortex. I. Evidence for monaural directional cues. J Neurophysiol 70: 492–511

Samson FK, Barone P, Clarey JC, Imig TJ (1994) Effects of ear plugging on single-unit azimuth sensitivity in cat primary auditory cortex. II. Azimuth tuning dependent upon binaural stimulation. J Neurophysiol 71: 2194–2216

Schreiner CE, Cynader MS (1984) Basic functional organization of second auditory cortical field (AII) of the cat. J Neurophysiol 51: 1284–1305

Semple MN, Kitzes LM (1993a) Binaural processing of sound pressure level in cat primary auditory cortex: evidence for a representation based on absolute levels rather than interaural level differences. J Neurophysiol 69: 449–461

Semple MN, Kitzes LM (1993b) Focal selectivity for binaural sound tuning. J Neurophysiol 69: 462–473

Taniguchi I, Nasu M (1993) Spatio-temporal representation of sound intensity in the guinea pig auditory cortex observed by optical recording. Neurosci Lett 151: 178–181

Taniguchi I, Horikawa J, Moriyama T, Nasu M (1992) Spatio-temporal pattern of frequency representation in the auditory cortex of guinea pigs. Neurosci Lett 146: 37–40