DEVELOPMENT OF TONOTOPY, THRESHOLDS, LATENCIES AND TUNING IN THE MOUSE INFERIOR COLLICULUS

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The development of the size of the mouse inferior colliculus (IC), tonotopy, tone response thresholds, latencies and tuning of neurons was investigated between postnatal days 10 (first responses) and 20. During that time, the size of the IC remained constant in all 3 planes. At day 10, the first low-frequency responses without tonotopy could be recorded with microelectrodes from neurons in the rostral and central parts of the central nucleus sparing its caudal part. Between days 10 and 20, there was a constant increase of highest characteristic frequencies (CFs) measurable of neurons in the IC. Neurons at a given constant collicular depth showed a clear shift of CFs from low to high, that is, they were tuned to the higher frequencies the older the animals were. As in adults, response thresholds of neurons are the lowest in the center of the IC compared with other areas right from the beginning of responsiveness at day 10. Thresholds decrease rapidly until days 16-20 (depending on the CF of the neuron) adult level are reached. Response latencies decrease rapidly to adult levels at days 16-18. Broad frequency tuning curve (TC) shapes differentiate and adult diversity occurs which indicates presence of inhibition and summation in the pathway to or within the IC at that early age. The tip-lengths of the TCs increase faster than the sharpness of the tips. The developmental courses of the measured parameters are expressed by power functions. The time constants of these functions are used in the discussion of processes underlying the functional maturation in the auditory system of the mouse.

Introduction

The mouse auditory system has become a widely used model for the study of the development of hearing. The beginning of the responsiveness to sound stimuli at postnatal day 10 and the end of the auditory-threshold development measured behaviorally at about day 18 are only 8 days apart (1). We may suggest that during these days many auditory functions such as resolution and encoding of sound spectra, intensity and direction become adult-like. Since the auditory system has so many levels of processing between the cochlea and centers relevant for sound recognition, it is important to know the developmental time courses at all levels in order to understand the contribution of these levels to the whole picture of improvement of function. Several behavioral activities to acoustical stimulation seem to be mediated at the level of the midbrain or below (2,3,4). Thus, strategies of coding auditory information in the IC may directly show up in behavioral responses. Although neuronal activities in the IC of young mice have been obtained before (5,6,7,8) they do not allow, however, a sufficiently precise determination of time course of development, which is necessary for a meaningful mathematical treatment of the data.

Materials and Methods

Seven house mice each of the following postnatal ages were used : 10, 11, 12, 13, 14, 15, 16, 18 and 20 days. The day following the night of birth was counted as postnatal day 1. Within a sound-proof and anechoic room neurons were recorded with steel electrodes from the right IC. Single unit or multi-unit responses were recorded in predetermined depths. At every depth the characteristic frequency and the absolute threshold of an excitatory response were determined audiovisually as that frequency and sound pressure level (SPL re. 20 μ Pa). The excitatory frequency TC was determined and the latency from tone burst onset to the first spike was measured 10 dB and 50 dB above threshold at CF. At the end of the recording session, frontal frozen sections (20 μ m) of the unfixed brain were made and stained with Cresyl violet for measuring the size of the IC. Electrode tracks were reconstructed from electrolytical lesions after histological reconstruction.

A formal expression of developmental growth can be obtained by growth functions of the types (9)

$$L_t = L_a \pm A e^{-t/b} \quad (L_t = \text{value of a measure at a certain age; } L_a = \text{adult value at the end of the development; } b =$$

time constant; A = scaling factor) introduced in this study leads to comparisons of time constants of various developmental trends in centers of the auditory pathway and thus to a common basis for the evaluation of the anatomical and physiological background of auditory maturation.

Results

The size of the IC remained constant in all 3 planes (rostrocaudal, mediolateral and dorsoventral) between postnatal ages of 9-20 days. Hence, frequency maps from the various postnatal ages can directly be compared without factors of adjustment for size or shape differences necessary, (fig. 1). At day 10, the first low-frequency responses without tonotopy could be recorded from neurons in the rostral and central parts of the central nucleus sparing its caudal part, very medial portions, the medial part of the central nucleus, the dorsal cortex and the lateral nucleus. Then, an extension of the frequency responsiveness occurred towards (1) the caudal pole which was reached by about day 14, (2) the dorsal surface reached between days 12 and 14, (3) the ventral border of the IC reached by about day 15. The high-frequency nucleus of the IC, which represents the medial part of the IC, remained unresponsive to tones up to day 13. Between days 10 and 20, there was a constant increase of highest CFs measurable of neurons in the IC. During that time, lowest measurable CFs remained rather constant. Neurons at a given constant collicular depth of more than 400 μm showed a clear shift of CF from low to high, that is, they were tuned to the higher frequencies the older the animals were, (fig. 1).

A comparison of threshold averaged from the center of the IC with those from the other areas shows that, irrespective of CF and ages, the most sensitive neurons are most often located in the center of the IC, where they have up to 40 dB lower average threshold compared to medial, lateral, caudal or rostral areas, (Fig. 2). The decrease of the thresholds can be described by the following functions calculated (by a regression analysis after linearization of the data) from the data of the center of the IC : CF-range 4-6kHz : $L_t = 17 + 12 \times 10^3 e^{-1/2.2}$; 14-16 kHz : $L_t = -5 + 9 \times 10^3 e^{-1/2.3}$; 28-32 kHz : $L_t = -1 + 7.5 \times 10^3 e^{-1/2.6}$. All functions express statistically significant correlations ($p < 0.01$). The time constants of the threshold developments in the medial, lateral, caudal or rostral parts of the IC are similar to those calculated from the center.

The development of tone response latencies was measured 10 dB and 50 dB above threshold at CF. Latencies decrease continuously from the beginning of the responsiveness up to about postnatal day 16. The most rapid decrease occurs between days 12 and 13. Although latencies of neurons with CFs above 15 kHz are, in general, shorter than those of low frequency neurons at both supra-threshold levels, significant differences ($p < 0.005$, U-test) occur only at postnatal days 11 and 12, when measured 10 dB above tone response threshold. The latency development measured 10 dB above tone response threshold for neurons with $CF \leq 15$ kHz can be expressed by the function: $L_t = 14 + 1.95 \times 10^3 e^{-1/2.28}$.

At day 10 and 11, most TCs are characterized by a steep slope to the high-frequency side and a shallow slope to lower frequencies. At these ages only a few curves show a tip around CF. The tip length however, is maximally 10 dB. From day 12 onwards, most TCs have a marked tip region. Also, their shapes become rather variable including very broad curves, curves with very steep slopes at both flanks, tilted curves, closed curves and complex ones with two tip areas. The sharpness of tuning near the tip of the TC can be expressed by the $Q_{10\text{dB}}$ -value (CF divided by the bandwidth 10 dB above the absolute threshold). A regression analysis reveals that $Q_{10\text{dB}}$ -values increase significantly with increasing CF for age groups 11, 12, 15+16 and 18+20 days ($P < 0.05$). For 13+14 days old animals, the correlation between $Q_{10\text{dB}}$ -values exists only as a trend because the significance level of $p < 0.05$, is just not reached. Figure 3 shows the average increase of the $Q_{10\text{dB}}$ -values calculated for a CF-range of 6 - 20 kHz to which animals of all ages contributed most TCs. Although the variability is large $Q_{10\text{dB}}$ -values at 11 and 12 days are significantly different from those of 14, 15, and 16 days ($p < 0.01$, U-test). The increase follows the function (days 18 + 20 not included in the calculation) : $L_t = 5.8 - 175 e^{-1/3.0}$.

Discussion

Our data show that all presently investigated response features of the IC undergo systematic changes during the postnatal development. Tonotopic representation changes, thresholds and latencies decrease, TCs shapes become diverse and the overall sharpness of tuning increases. After the onset of responsiveness a series of changes of frequency representation took place. For example, the progression of frequency responsiveness ventralwards in the medial part and the increase of CFs with increasing age does not mean that the CF-increase is accomplished just by the initiation of activity in more and more ventral parts of the IC as found by Schäfer and Rübsamen (10). Our present data and those of the kitten (11) demonstrate a clear shift of CF from low to high for a given constant depth (dorsoventral position) in the IC. Such a shift is seen for all

Fig. 1 : Drawing of frontal sections of the IC. The section is approximately from the middle of the nucleus (rostrocaudal axis) 500 to 700 μ m from the caudal pole. Modifications of frequency representation can be observed by isofrequency lines or fields across animals of different ages.

Fig. 2: Development of average tone response thresholds for neurons with CFs in the range of 14-16 kHz. Mean from neurons in the central, medial, lateral, caudal and rostral part of the IC are shown separately. Standard errors are indicated for the means from the center of the IC. These values are approximated by regression lines (calculated after linearization of the data) the parameter of which are given in the text. Adult values (x) are from Ehret and Moffat (23).

Fig. 3 : Change of average Q_{10dB} -values with increasing age of the animals. Means and standard deviations are shown. At 18+20 days only five values could be averaged which probably was too small number for yielding a meaningful result. The Q_{10dB} increase is approximated by a regression line, the parameter of which are given in the text. The adult value is averaged from data of Ehret and Moffat (23).

depths of more than 400 μm . That is the frequency-place relationship in the IC, including the central nucleus and its medial part has dynamic properties. Neurons at a given place first respond best to rather low frequencies and then to increasing higher frequencies. This is an important result because it shows that the dendritic laminae and ascending fibers in the central nucleus of the IC are tuned to different frequencies at different postnatal days. Such expansion of the CF-range towards higher frequencies were observed in all mammalian midbrain from which respective recordings exist (5,7,8,12,13). This feature, therefore, seems to be a general one of auditory maturation and may have importante consequence for auditory perception (5).

The tone-reponse threshold measured in mice by Shneron and Willott (7) can hardly be compared with our data because they relate to periods not to single days. The trend of threshold decrease, as described here, is present also in this study. Sanes and Constantine-Paton (6) determined click-evoked potential thresholds in the IC of mice. There values are higher than those of single neurons and the threshold decrease much faster with time constant of 1.5 compared to our data. One important reason for this discrepancy concern the synchronization of neuronal responses at the onset of the click that may be very important in immature animals. The time constant of the latency development of low-frequency neurons measured 10 dB above threshold is somewhat larger than that of high-frequency neurons (2.28 vs. 1.79). This may indicate a faster development of the input to the high frequency neurons of the central nucleus of the IC, because they receive information from the mid-basal part of the cochlea the maturation of which is the most advanced at the beginning of hearing (14, 15, 16).

The frequency selectivity expressed by the tip of the TC improves mainly by lengthening and narrowing the tip. One could argue that the tip-length should critically depend on the response threshold at CF. The lower the threshold is the longer could be the tip of the TC. The development of the TCs in the auditory nerve and cochlear nucleus seems to follow this rule (17,18,19,20,21) and on the first view this relationship seems to hold also in the IC of the mouse. A close study on the time constants reveals, that the length of the tip increases faster with the major development between day 12 and 14 compared to the threshold decrease at any CF. Such a maturation of TCs running ahead of the threshold development has been observed already in the auditory nerve of kittens (22), and may be refined in higher centers by mechanisms of lateral inhibition coming into play.

In summary, time courses of the development of auditory features such as thresholds, latencies and tuning differ because of differences in the methods of measurements and/or physiological mechanisms. A formalization of the developmental courses leading to time constants may help to direct the attention to such differences so that true mechanisms and factors of maturation may easier be discriminated from influence of methodology.

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