3rd Quarterly Progress Report
April 1 to June 30, 2001

Neural Prosthesis Program Contract N01-DC-0-2108

Protective Effects of Patterned Electrical Stimulation on the Deafened Auditory System

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ABSTRACT

Previous reports from this Contract research have demonstrated that the temporal resolution and response latencies of neurons in the inferior colliculus (IC) can be markedly altered by chronic electrical stimulation delivered through a cochlear implant. Temporal resolution, defined as the capacity of IC neurons to phase lock or follow electrical pulse trains delivered at increasing frequencies, is significantly increased in neonatally deafened cats following a period of several months of chronic stimulation (Snyder et al., J. Neurophysiol., 73:447-467, 1995). Further, subsequent studies showed that the magnitude of this effect is dependent upon the specific frequency(ies) of the chronically applied electrical signal. Low-frequency stimulation (e.g., 30 pps unmodulated pulse train) maintains temporal resolution comparable to that seen in normal control subjects, whereas stimulation with more temporally challenging signals which approach the normal upper limit of IC responses (e.g., 300 pps carrier amplitude modulated at 30 Hz, or an analogue cochlear implant processor) results in a marked increase in temporal resolution and significantly shorter first-spike latency (Vollmer et al., J. Neurophysiol., 82: 2883-2902, 1999). This Quarterly Progress Report extends these previous investigations by presenting preliminary data from 7 animals chronically stimulated with even higher frequency signals using carriers ≥500 pps. The findings in these subjects are compared to previously published data from acutely deafened 'control' animals and to animals stimulated with the 300 pps/30 AM signal.

INTRODUCTION

The impressive speech reception ability enjoyed by many cochlear implant (CI) recipients using the latest prosthetic devices has been attributed to advancements in sound processing strategies -- in particular, the use of amplitude modulated, interleaved pulse trains used to encode acoustic signals in contemporary CIS processors. Since these newer, high-pulse rate CIS processors appear to provide additional benefit to CI users and since temporal resolution has been shown to be important for the perception of pitch, prosody and speech (e.g., Eddington et al. 1978, Shannon 1983, Wilson et al. 1991), we have been particularly interested in studying the capacity of central auditory neurons to encode and resolve the temporal characteristics of electrical stimuli. We hypothesize that the ability of the central auditory system to respond to specific temporal features of electrical signals is likely to be particularly important to an understanding of the effects of experience (plasticity) on auditory processing and the success of 'electrical hearing'.

We have previously reported that the temporal features of chronically applied intracochlear electrical signals critically influence the temporal resolution of single IC neurons. The maximum following frequency or Fmax and the response latencies to electrical pulse trains was measured in isolated single neurons arrayed across the cochleotopically organized inferior colliculus (IC) (QPR#5, 1995; Snyder et al., J. Neurophysiol., 73:447-467, 1995; Vollmer et al., J. Neurophysiol., 82: 2883-2902, 1999). Specifically, chronic electrical stimulation of the cochlea in neonatally deafened animals using low-frequency signals (30 pps, 80 pps) *maintains* the temporal resolution of IC neurons, such that the average temporal following for neurons is similar to that seen in normal control subjects. In contrast, several months of chronic stimulation with higher-frequency stimuli specifically designed to be temporally challenging to the central auditory system results in a significant *increase* in the average temporal resolution of IC neurons. In this temporally challenging stimulation group, neurons in the IC exhibited a significantly higher mean Fmax and significantly shorter median

response latencies as compared with neurons in adult, acutely deafened 'control' subjects. The temporally challenging chronic stimulation applied in this experimental group was either: 1) a continuous train of biphasic electrical pulses delivered at a carrier rate of 300 pps and sinusoidally amplitude-modulated (SAM) at 30 Hz (100% modulation depth); or 2) input from an analogue processor (SP) that transduced sounds in the animal's environment (band-pass filtered from 250 Hz to 3 kHz). Interestingly, the increase in temporal resolution observed following temporally challenging stimulation was restricted to neurons within the central nucleus of the IC (ICC), and no significant effect of stimulation was recorded in the external nucleus (ICX).

Electrophysiological studies in normal hearing animals using acoustic stimulation have shown that most IC neurons follow stimulus frequencies up to 100 Hz and some can respond to higher frequencies up to about 300 Hz, but very few neurons (<0.1%) can resolve frequencies $\geq$500 Hz (e.g. Batra et al 1989, Langner and Schreiner 1988). Results to date from electrophysiological studies of intracochlear electrical stimulation in deaf animals have shown parallel results. Very few IC neurons respond at all to electrical pulse trains with carrier frequencies $\geq$300 pps (Snyder et al. 1995, 2000), even following several months of experience with temporally challenging chronic stimulation (Vollmer et al. 1999). Moreover, in human psychophysical studies on rate pitch discrimination, Shannon (1983, 1985, 1992) has reported that CI subjects can discriminate repetition or modulation frequencies only up to ~300 pps.

These findings raise the question of how the central auditory system can make use of the high rate signals delivered by current CIS processors. Can the central auditory system, over time, adapt to these faster repetition rates and acquire the capacity to respond to and encode higher frequency signals? How does *chronic stimulation* at frequencies exceeding the normal Fmax of IC neurons influence the temporal resolution of these neurons? To address these questions, we conducted additional experiments using chronic stimulation with high frequency carriers ≥500 pps and examined the functional consequences of these signals on temporal processing

of IC neurons. Preliminary data from 7 neonatally deafened cats that underwent chronic stimulation with higher frequency signals are reported in this QPR and compared to data from acutely deafened adult "control" subjects and to neonatally deafened cats that received chronic stimulation with 300pps/30Hz SAM.

METHODS

Table 1 documents the chronic stimulation histories of the neonatally deafened experimental animals included in this report. Results were obtained from six animals that received several months of chronic stimulation using a temporally challenging stimulus comprised of a continuous 300pps carrier comprised of 200 μsec/phase biphasic pulses that were sinusoidally amplitude modulated (100% modulation depth) at 30Hz ("Temporally Challenging Stimulation Group"). A second group of seven animals in the "High Frequency Stimulation Group" received chronic stimulation either with: 1) a continuous, invariant electrical signal with a carrier rate of 800 pps that was sinusoidally amplitude modulated at 20 or 60 Hz; or 2) carrier frequencies that were systematically stepped from 100 pps to 800 pps throughout the stimulation period. In the latter case, stimulation was applied for one week using one of four temporally challenging signals in the following sequence: 100 pps (unmodulated), 300 pps/30 Hz, 500 pps/40 Hz, and 800 pps/50 Hz. This sequence was repeated until the completion of chronic stimulation periods.

Animals in both experimental groups were deafened neonatally by the systemic administration of aminoglycosides beginning immediately after birth. Profound hearing loss (>108 dB SPL) was confirmed by auditory brainstem response (ABR) testing to clicks. At 7-11 weeks of age, the animals were implanted with a scala tympani electrode array in the left cochlea. Implants consisted of four ball-shaped electrode contacts (200-300 mm in diameter) labeled 1 through 4 from apical to basal cochlear locations. The electrodes were arranged as two bipolar offset-radial pairs (apical pair 1,2; basal pair 3,4) with 1 mm separation between electrodes comprising a pair and 4 mm between the two pairs. A percutaneous connector allowed direct electrical connection to the electrodes. Chronic stimulation was applied for 4 hr/day, 5 days/wk, over periods ranging from 14 and 38 weeks. The average stimulation period was 30.2±6.3 (SD) weeks for the 300/30 group and 25.1±6.5 (SD) weeks for the ≥500 pps groups. The difference in the stimulation periods was not significant ($p>0.1$, Student's t-test). Some animals were chronically

stimulated with a single channel (apical electrode pair 1,2). Other animals received two-channel stimulation using both electrode pairs 1,2 and 3,4. The two channels were stimulated either concurrently (K143) or alternating from day to day (alt. d.; K107) or alternating two hours/day for each electrode pair ('alt. hr.'; Table 1).

Fourteen acutely deafened adult cats with prior normal auditory experience served as 'controls'.

Table 1: HISTORIES OF EXPERIMENTAL ANIMALS

Cat #	Stim.Period (weeks)	Channel(s)	Stim. Frequency (pps)	Fmax (ICC) (pps)
	Temporally Challenging Stimulation Group (300 pps/30 Hz)			
K89	27	single channel (1,2)	300/30	145
K91	31	single channel (1,2)	300/30; Beh	151
K92	24	single channel (1,2)	300/30	116
K99	37	single channel (1,2)	300/30; Beh	140
K102	38	single channel (1,2)	300/30; Beh	129
K143	24	2-channel (concur.)	300/30	108
	High Frequency Stimulation Group (≥500pps)			
K105	28	single channel (1,2)	800/20	114
K106	32	single channel (1,2)	800/20	122
K107	24	2-channel (alt. d.)	800/60	76
K130	22	2-channel (alt. hr.)	var: 100-800/50	105
K133	33	2-channel (alt. hr.)	var: 100-800/50	102
K134	14	2-channel (alt. hr.)	var: 100-800/50	76
K136	23	2-channel (alt. hr.)	var: 100-800/50	102

Table1: conc.=concurrent stimulation of electrode pairs 1,2 and 3,4 (4 hrs/d);
alt. d=stimulation delivered on electrode pair 1,2 or 3,4 on alternating days (4 hrs/d);
alt. h= alternating stimulation, 2 hrs/d for each electrode pair 1,2 and 3,4.

In final electrophysiological experiments, the IC contralateral to the implant was surgically exposed under barbiturate anesthesia. Using tungsten microelectrodes, multi- and single neuron thresholds were "mapped" across the IC using biphasic pulses (0.2 ms/ph; 2-5 pps) and sines (3 cycles of 100 Hz) recorded at 100 µm intervals perpendicular to the isofrequency laminae of the IC (Snyder et al. 1990, 1995, Vollmer et al.1999). Thresholds were plotted as a function of IC depth, generating functions which were typically w-shaped (Fig. 1). These threshold functions are called "spatial tuning curves". The high threshold region between the two regions of minimum thresholds indicates the border between the two subnuclei of the IC and allows neurons to be assigned to either ICX or ICC.

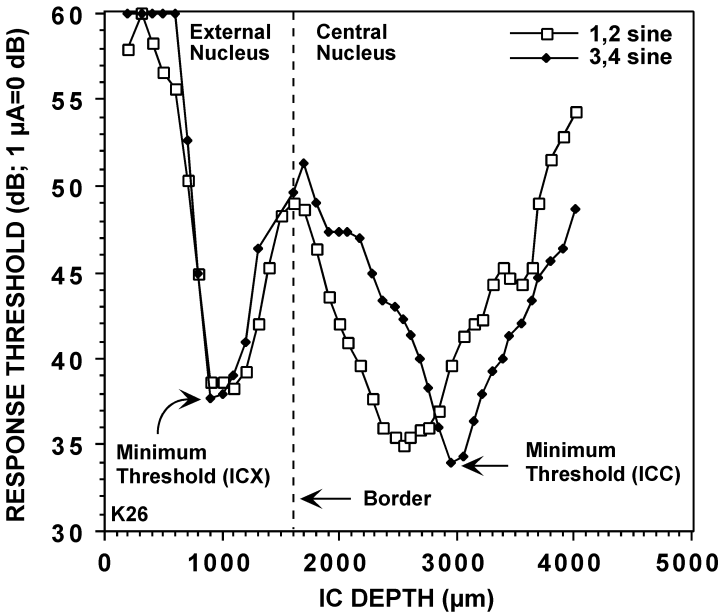

Figure 1. Spatial tuning curves for sine thresholds for stimulation of the apical electrode pair 1,2 (open symbols) and basal electrode pair 3,4 (closed symbols) plotted as a function of IC depth. High threshold region indicates border between external and central IC nuclei (dashed line).

Single neuron responses were recorded at any depth within the IC where they could be isolated from the stimulus artifact at a stimulus intensity of 2-6 dB above threshold (calibration: 0 dB=1µA peak to peak, µApp). To investigate the influence of the chronically applied stimuli on temporal processing, the activity of single neurons was recorded in response to: 1) pulse trains of increasing frequencies (beginning at 10

pps, increments of 5-10 pps) until the neuron failed to respond or responded to the stimulus with only an onset spike burst; and 2) amplitude modulated pulse trains of increasing modulation and carrier frequencies (i.e., to signals including those used for chronic stimulation in the 300 pps/30 Hz and ≥500 pps groups). The frequency to which the neuron exhibited significant phase locking to the stimulus ($p<0.01$, Raleigh test) was determined and defined as the maximum following frequency (Fmax). In addition, first spike latencies were measured using responses to stimulus frequencies of 20 pps.

RESULTS

1. AVERAGE MAXIMUM FOLLOWING FREQUENCIES TO UNMODULATED PULSE TRAINS

Figure 2 summarizes the average Fmax in the ICX and the ICC for the normal control subjects and for the two experimental groups. In all three groups, responses of neurons in the ICX had significantly lower Fmax than those in the ICC ($p<0.01$; Student's t-test, unpaired). In the ICX the highest temporal resolution was recorded in animals in the temporally challenging stimulation group, chronically stimulated at 300pps/30 Hz AM, in which the average Fmax was 77.3 pps (±8.3, SE; number of neurons 'n'=35). In the high frequency group stimulated with carrier frequencies ≥500 pps the mean Fmax for the ICX was 62.6 pps (±5.3, SE; n=5), a value which was very similar to that of the control animals which had a mean Fmax of 58.3 pps (±6.1, SE; n=28). However, statistical comparisons failed to demonstrate any significant difference among the three groups.

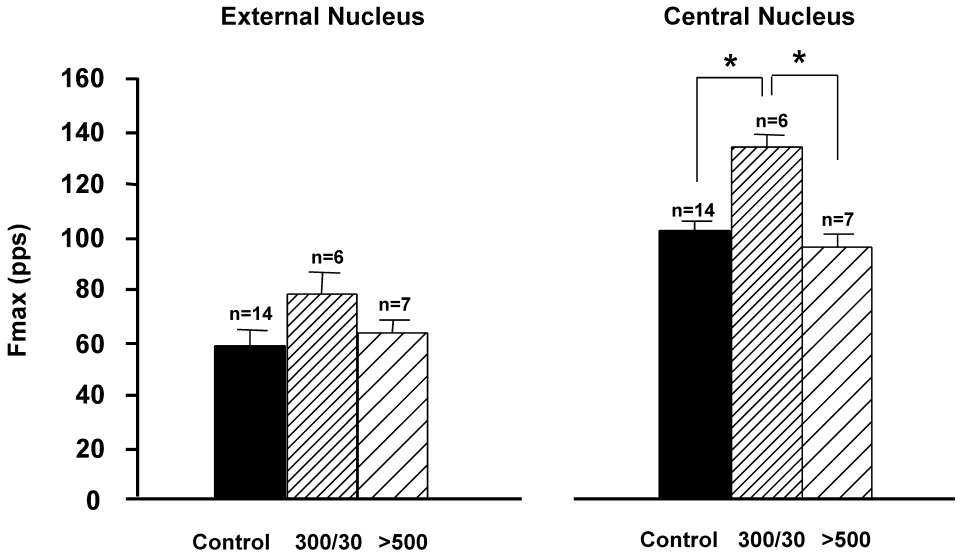

Figure 2: Average Fmax for experimental groups separately displayed for the two subnuclei of the IC. Asterisk (*) indicates statistical significance ($p<0.01$; t-test). N=number of animals. Error bars=standard error (SE).

The analyses of single neuron responses in the ICC, showed that Fmax was significantly higher than in the ICX for all groups. The overall range of Fmax values in the ICC was similar in all three groups with values ranging from about 10 to about 330 pps. However, the average values of the distributions showed significant differences. In control animals the mean Fmax of ICC neurons (n=226) was 101.6 pps ±3.8 (SE). Following chronic stimulation with carrier frequencies of 300 pps, ICC neurons (n=142) demonstrated a highly significant increase in average Fmax to 132.9 pps ±4.7 (SE). In contrast, following chronic stimulation with carrier frequencies ≥500 pps the average Fmax of ICC neurons (n=160) was only 95.8 pps ±4.5 (SE) pps, a value that was virtually identical to (and not significantly different from) the Fmax for unstimulated control animals. Statistical comparisons demonstrated that the mean Fmax of the high frequency group stimulated at ≥500 pps was significantly lower than that from animals chronically stimulated with a 300 pps carrier.

2. MEDIAN LATENCIES TO UNMODULATED PULSE TRAINS

The median values and quartile deviations (Q) of first spike latencies for the three experimental groups are displayed in Figure 3. Overall, responses from neurons in the ICX of all three groups had significantly longer latencies than those in the ICC ($p>0.01$, Mann-Whitney U test). ICX neurons (n=30) from control animals had a median latency of 8.9 ms ±1.8 (Q), neurons from animals stimulated with 300/30 AM (n=35) and ≥500 pps (n=56) had median latencies of 8.4 ms ±1.2 (Q) and 9.0 ms ±1.3 (Q), respectively. There were no significant differences among these median latencies of ICX neurons recorded in the three different groups. In the ICC, however, significant differences in latency were observed among the experimental groups. In the control animals the median latency of ICC neurons (n=232) was 6.9 ms ±0.8 (Q). In contrast, neurons from animals stimulated with the temporally challenging signal (300 pps/30

Hz AM) had significantly shorter latencies with a median of 5.7 ms ±0.9 (Q) (n=146). In addition, neurons recorded in the high frequency group chronically stimulated with carrier frequencies ≥500 pps had a significantly longer median latency (7.0 ms±1.4 (Q); n=161) when compared to either the 300 pps/30 Hz AM group or the control group. It should be noted that although the difference between the median latency values for the high frequency stimulation and normal groups was very small (7.0 vs 6.9 ms), the latency distributions for the two groups were different (Fig. 4). Moreover, the *mean* latency for the normal group was 6.81 ms as compared to 7.4 ms for the high frequency stimulation group. Thus, statistical analysis demonstrated a significant difference between these two groups.

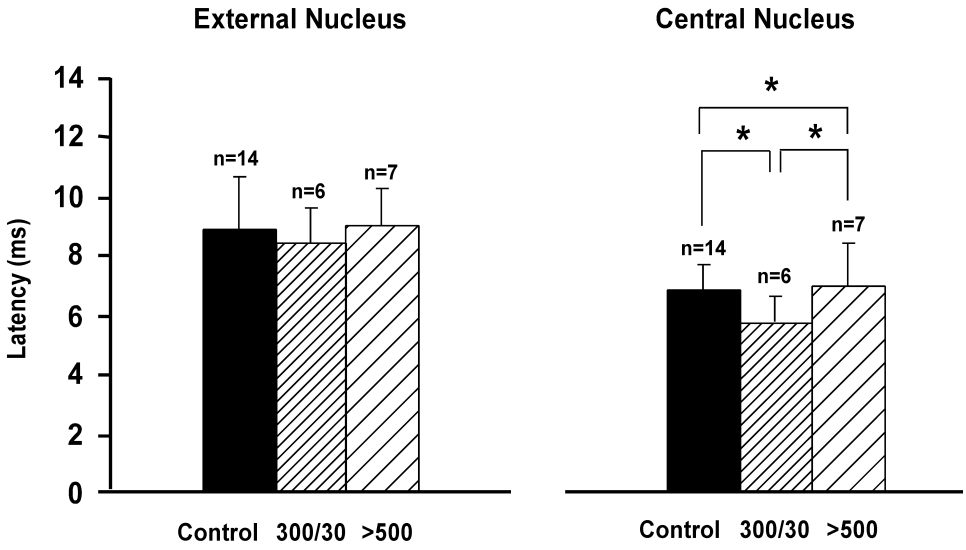

Figure 3: Median latencies for experimental groups separately displayed for the two subnuclei of the IC. Asterices (*) indicate statistical significance ($p<0.01$; Mann-Whitney U test). N=number of animals. Error bars= quartile deviation (Q).

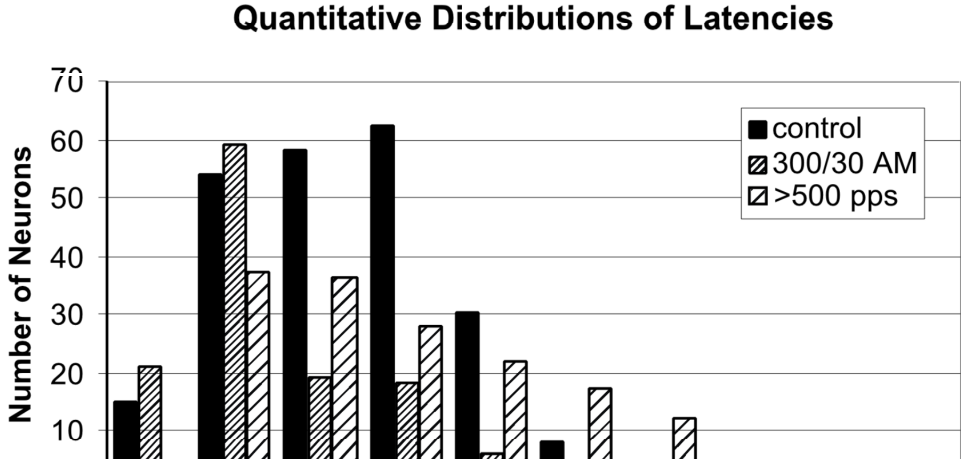

Figure 4: Distributions of first spike latencies for ICC neurons are shown individually for the three experimental groups

The above Fmax and latency data support the previously reported finding that chronic electrical stimulation with carrier frequencies that are within the upper range of 'normal' Fmax of ICC neurons (~300 pps) results in improved temporal resolution (higher Fmax and shorter response latencies) in the population of ICC neurons (Vollmer et al. 1999). The results presented here include data from one additional cat in this temporally challenging stimulation group. Moreover, the current results from the high frequency stimulation group suggest that chronic stimulation with carrier frequencies that exceed the 'normal' range of Fmax may either *maintain* or *restore* the temporal resolution of ICC neurons in these neonatally deafened animals to "normal" values, but fails to improve temporal resolution beyond the normal limit. Moreover, the longer latencies observed in animals stimulated at >500 pps may suggest a relative decrease in temporal resolution as compared to the other groups.

3. SINGLE NEURON RESPONSES TO AMPLITUDE MODULATED PULSE TRAINS

In order to interpret the difference in the Fmax of ICC neurons observed in the two chronically stimulated experimental groups, it is important to consider how these neurons respond to SAM. We have previously reported on the responses of single neurons in the IC to SAM stimuli of increasing carrier (100-1000 pps) and modulation

(usually 8-60 AM) frequencies (Snyder et al., 2000) and compared the Fmax to the maximum modulation frequency that these neurons followed. Exemplary responses of two IC neurons, one low resolution neuron and one high resolution unit, to unmodulated (left columns) and SAM pulse trains (two right columns) are presented in Figures 5 and 6.

Figure 5 shows the responses of neuron "A" which has relatively low temporal resolution. The Fmax of this neuron responding to unmodulated pulse trains (left panel in Fig. 5) is 30 pps. In the right two panels of the figure, the responses of the same neuron to SAM stimuli with carrier frequencies of 100-400 pps are displayed. The modulation frequencies in these examples are 8 Hz and 24 Hz. In none of these recordings does the neuron phase lock to the carrier rate of the signal, instead it phase locks strongly to the modulation frequencies, which are much lower and fall well within the range of frequencies the neuron follows in its responses to unmodulated pulse trains. Thus, the responses of this neuron are relatively independent of the carrier frequency and are similar to the responses of a neuron to unmodulated pulse trains of 8 Hz and 24 Hz.

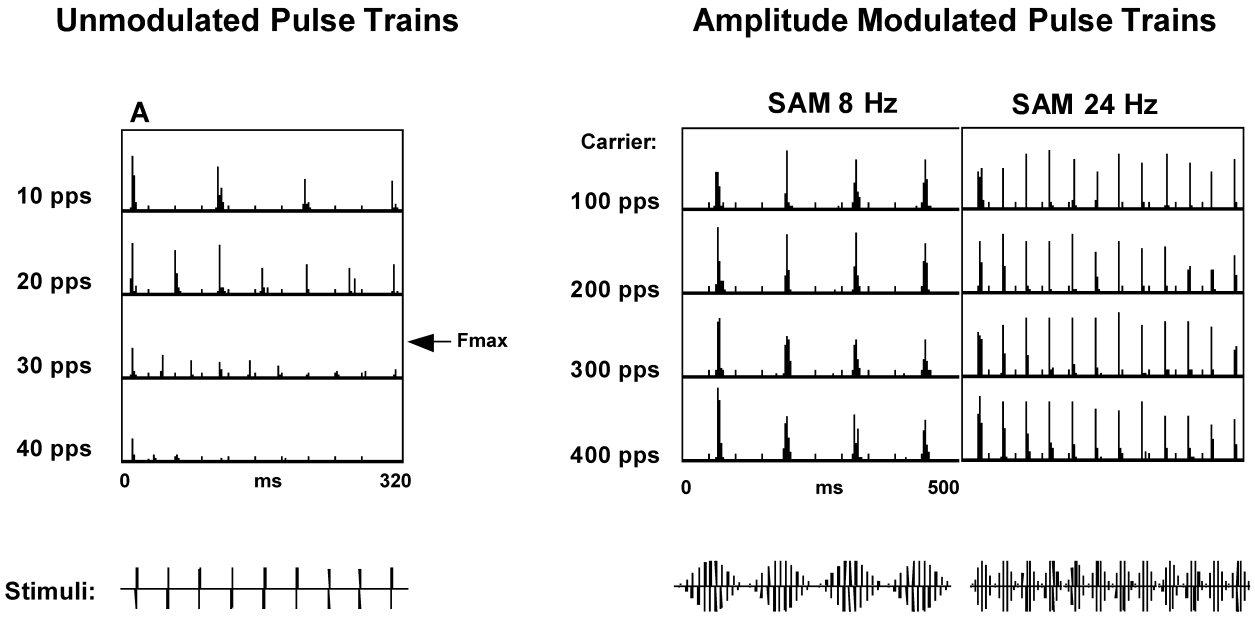

Figure 5: Poststimulus time histograms (PSTHs) for responses of a single neuron, denoted neuron "A," with low temporal resolution (Fmax=30 pps, K104) responding to unmodulated

pulse trains (left panel) and SAM pulse trains of increasing frequencies (right panels). Examples of the electrical stimuli applied in each series are illustrated below.

Figure 6 illustrates the responses of a second neuron, denoted 'B', to unmodulated (left) and SAM (right) pulse trains. The temporal resolution of this neuron is clearly much higher than that of neuron "A" with an Fmax slightly greater than 300 pps. In responses to SAM at the lower modulation frequency (8 Hz), neuron B phase locks to carrier frequencies up to 300 pps ($p<0.01$, Raleigh test), i.e., to carrier frequencies that correspond to the neuron's Fmax to unmodulated pulse trains. That is, within each of the 3 spike clusters which correspond to the maxima in the 8 Hz envelope of the stimulus, there are a series of peaks which are time-locked to the carrier rate of the specific stimulus. At a higher modulation frequency of 24 Hz, however, neuron B displays strong phase-locked responses only to the modulation frequency of the stimulus. This exemplary high resolution neuron responds relatively independently of the carrier rate (although peaks in the fine structure representing the carrier frequency are occasionally visible in responses to carriers up to 300 pps). Thus, the response to the 24 Hz SAM signals are again somewhat similar to the responses of a neuron to an unmodulated pulse series at 24 pps.

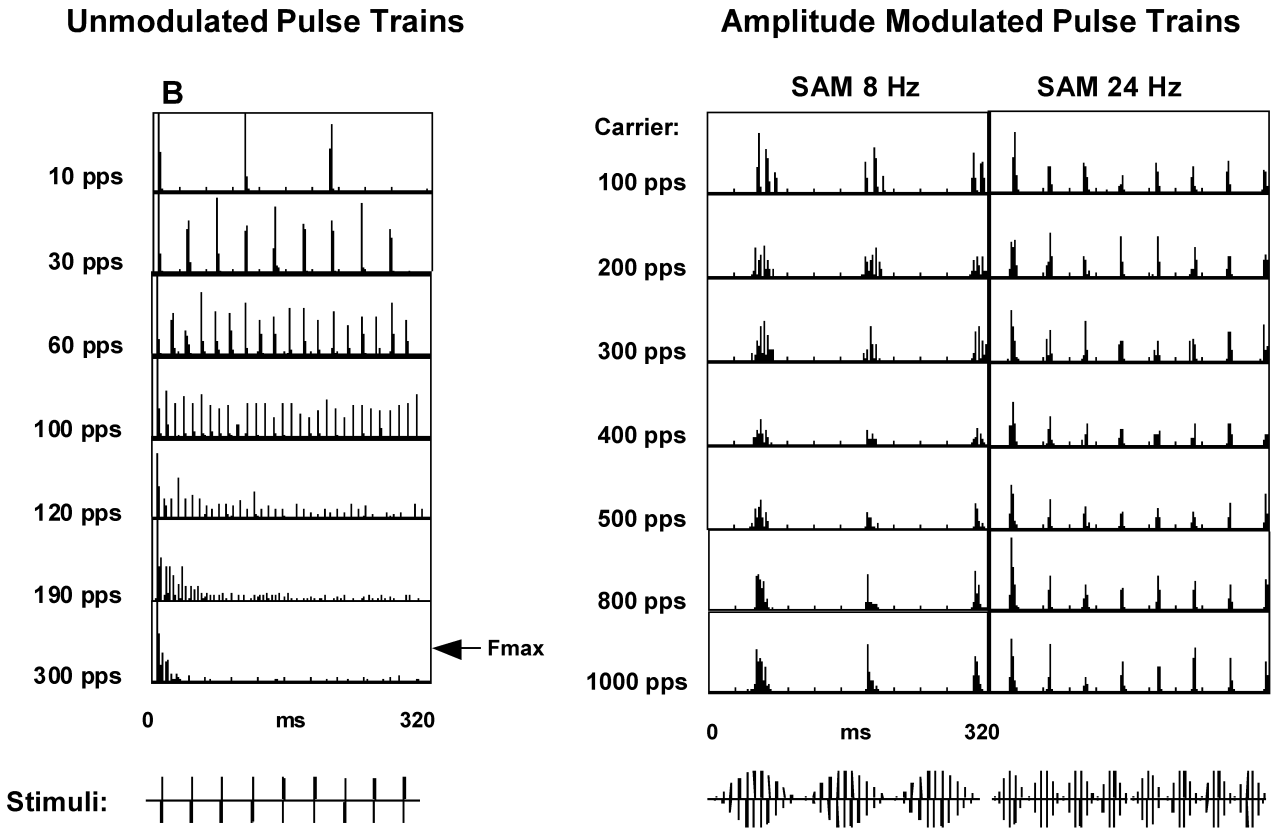

Figure 6: Poststimulus time histograms (PSTHs) for responses of a single neuron ('B') with high temporal resolution (Fmax=~300 pps; CH611) to unmodulated and SAM pulse trains of increasing frequencies. Examples of the used stimuli are illustrated below.

SUMMARY AND CONCLUSIONS

These preliminary findings demonstrate that the range of Fmax of ICC neurons is similar in all experimental groups regardless of chronic stimulation history and relative experience with the applied electrical signals (i.e., about 10 to 330 pps or the 'normal' range). However, the mean values of the distributions of Fmax for the three different groups showed significant differences. The results suggest that chronic stimulation with carrier frequencies near the high frequency end of the normal range of Fmax (300 pps) resulted in a significant improvement in the temporal resolution of ICC neurons (higher Fmax, shorter latencies). In contrast, chronic stimulation with carrier frequencies that

exceed the normal range of Fmax (≥500 pps) did not significantly alter the frequency following ability of ICC neurons from that seen in normal, unstimulated control subjects (virtually identical average Fmax). That is, such higher frequency stimulation fails to improve temporal resolution but does maintain relatively normal temporal following in the IC of these neonatally deafened animals. Moreover, latencies of ICC neurons from animals stimulated with carrier signals ≥500 pps are significantly longer than those from control animals, suggesting a degradation in temporal resolution in this group.

Electrophsyiological data recorded from individual IC neurons responding to SAM signals suggests that phase locking to the SAM carrier is usually limited to frequencies that are equal to or less than the specific unit's Fmax for unmodulated pulse trains. At higher carrier frequencies, IC neurons generally respond exclusively to the relatively low modulation frequency (8-60 Hz) and responses appear relatively unchanged with any further increase in carrier frequency.

The above observations suggest that for stimulation with carrier frequencies that exceed the normal range of Fmax the modulation envelope provides the dominant *temporal* information. The data suggest that pulse rates much above about 350 pps would not contribute additional information to the fine structure of neuronal responses within the central auditory system, at least at the level of the auditory midbrain. Thus, it is unclear how the very high pulse rates of contemporary CIS processors contribute to information transfer to central auditory neurons.

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Work Planned for the Next Quarter

1) Several members of the UCSF group will attend the international Conference on Implantable Auditory Prostheses to present results of research conducted for this Contract. Dr. Leake will present an invited paper. Maike Vollmer, Steve Rebscher, Charlotte Moore, Julie Bierer, Ralph Beitel, Peter Wardrop and David Whinney will present posters. The 8 abstracts for these presentations are appended.

2) Chronic stimulation will be completed and the acute electrophysiological experiment will be conducted in the final animal in our "long-deafened" experimental series. This study is designed to examine the consequences of severe neural degeneration upon electrophysiological thresholds, dynamic range and the effects of chronic stimulation in this model of severe cochlear pathology.

3) Chronic stimulation will be continued in 2 animals in a new chronic series in which profound hearing losses are induced at 1 month of age using acute administration of kanamycin/ethacrynic acid, rather than the neonatal prolonged administration of neomycin used in most prior studies.

4) Additional quantitative analyses will be undertaken to examine the responses of neurons to unmodulated and SAM pulse trains with special attention to the relationships between Fmax for unmodulated and SAM carrier frequencies. The results will be analyzed for the different experimental groups, and since chronic stimulation differently affects temporal resolution in the ICX and ICC, the results will also be analyzed individually for the two subnuclei of the IC.