Chapter 2

Temporal aspects of the responses of auditory-nerve fibers to two-tone stimuli

Introduction

Two-tone suppression (2TS) is the phenomenon by which the responses to the characteristic frequency (CF) tone are suppressed by a second tone, the suppressor (SUP) tone. This phenomenon has been shown to exist at different stages of the auditory periphery: the basilar membrane (BM), the inner hair cell (IHC), and the auditory nerve (AN) fiber (i.e., mechanical, receptor and neural). It has been related to the "active" process, also referred to as "cochlear amplifier" (Dallos, 1992), within the cochlea. On the auditory nerve fibers, there have been many studies of 2TS since the first study by Galambos and Davis (1944) more than five decades ago. But most studies of 2TS (both experimental and model studies) seem to be restricted mainly to exploring reductions of discharge rate. Only a few studies concentrated on the temporal aspects of this suppression. However, in addition to our extensive knowledge of the rate aspects of two-tone suppression, the temporal aspects become increasing more important in our attempts to understand the 2TS phenomenon and the cochlear amplifier in general.

Among the limited number of temporal studies, different techniques were used. For example,

Javel et al. (1983) used synchrony (i.e., the synchronization to the CF tone) as a measure of the suppression strength, and Deng and Geisler (1985) looked at the change in the phase of the response to the CF tone induced by the suppressor. One particular approach, used in several studies, involves harmonically related CF tones and low-frequency suppressor tones, which allows one to see how the responses to the CF-tone are suppressed in a cycle by cycle manner by the suppressor tone (e.g. Sachs and Hubbard, 1981, Sellick et al., 1982 and Patuzzi et al. 1984a on AN; Patuzzi and Sellick, 1984 on IHC; Patuzzi et al., 1984b, Ruggero et al., 1992 and Rhode and Cooper, 1993 on BM). Patuzzi et al. (1984b) measured the responses of the basal region of the guinea pig BM, using a 33-Hz suppressor tone. They found that, at low suppressor intensities, the suppression occurs at the phase of maximum displacement of BM movement towards the scala tympani (ST), while at high suppressor intensities a second (and less pronounced) suppression phase appears, corresponding to the maximum displacement towards the scala vestibuli (SV). However, the relatively insensitive and nonlinear Mössbauer method prevented them from obtaining further details of the BM responses. Recently, with the advance of technology, several groups have begun to use a laser interferometer to study the BM responses. Using a displacement sensitive laser interferometer, Rhode and Cooper (1993) recorded from the hook region of the cats. They found a two-tone suppression pattern which generally agrees with that of the Patuzzi et al. (1984b), but also noticed a 0.1 ms lag when a 1000 Hz suppressor was used. In both studies, the time between the two phases of suppression was about half a cycle. Ruggero et al. (1992) used a velocity sensitive laser device to study the two-tone responses of the basal region of chinchilla BM, using 300-500 Hz suppressors. Although they found basically the same suppression patterns as the other investigators did (i.e., one suppression at lower intensities, also a second one at higher intensities), they observed, however, that the maximum suppression occurred on the phase when the BM was moving with maximum

velocity toward ST. The differences in these various studies have not been resolved yet.

At the IHC level, Patuzzi and Sellick (1984) found that both the AC and DC components of the CF-tone elicited receptor potentials of IHCs from basal region of the guinea pig were modulated in synchrony with a simultaneously applied low frequency suppressor tone (40 Hz). The suppression pattern was similar to those observed on BM by Patuzzi et al. (1984b).

Sachs and Hubbard (1981) were the first who studied 2TS on the auditory nerve fibers of cats using harmonically related stimuli. They found that, with 500 Hz and 100 Hz suppressors, there was usually one phase of suppression in the cycle of the suppressor tone at lower intensities, but there was also a second one when higher intensities were used. This basic pattern, which is the same as that revealed at the BM and IHC stages, was also confirmed by Sellick, Patuzzi and colleagues (Sellick et al., 1982; Patuzzi et al., 1984a) in the ganglion cells from the basal region of guinea pigs, using 40 Hz suppressor tones. In addition, Sellick et al. (1982), using CM recorded from ST as reference, found that the major suppression (which appears at lower intensities) was in phase with maximum BM displacement towards ST while the minor one (which appeared at higher intensities) was in phase with maximum BM displacement towards SV. Sachs and Hubbard (1981) also noticed that the major suppression was in phase with the phase of excitation to the suppressor tone alone. However, a close examination of the figures in their publication revealed some examples that depart from this general conclusion, especially in low-CF units (limited number).

Because of the discrepancies existing in the different studies and the fact that so far there has been no temporal study of 2TS from the apical region of the cochlea at any level using

harmonically related stimuli, we believe that more systematic studies in the basal as well as apical regions are needed at all levels of the system. It is the goal of this paper to provide a more systematic and thorough study at the AN level, using harmonically related CF and suppressor tones. Some of the data in this report have been presented previously (Cai and Geisler, 1994).

Methods

The animal preparation, sound delivery and recording system have been described in detail in Chapter 1. Briefly, the pinna of the anesthetized cat was removed. A silver wire electrode was placed near the round window through an opening on the bulla, to monitor the condition of the cochlea through the compound action potentials (CAP). The bulla was then re-sealed, with a thin plastic tube providing release of static pressure. The skull was opened in the posterior fossa area and the auditory nerve was exposed. A Trent-Wells hydraulic microdrive was mounted and fixed onto the skull, and a glass electrode (30-70 mΩ) was inserted in the nerve under visual guidance.

The stimuli were two harmonically related pure tones. The frequency of the suppressor (SUP) tone was always lower than fiber CF (with a frequency ratio no greater than 1/3), and it usually took one of the following values: 50, 100, 200, 500, 1000 and 2000 Hz. The frequency of the CF tone was adjusted slightly to ensure a harmonic relationship between the CF tone and the suppressor tone. The CF intensity was usually fixed at 10 to 30 dB above fiber threshold at CF. The suppressor intensity was varied over a range (e.g., from 70 to 95 dB SPL), usually in 5 dB steps. When a fiber was located by a frequency modulated

stimulus, a short paradigm in which two tones are presented simultaneously was used to make sure that 2TS exists. Then the paradigm shown in Fig. II-1 was used. The repetition time was 1000 ms and each stimulus lasted 500 ms. The suppressor was delayed by 200 ms so that there were three time windows corresponding to stimulation by CF-tone alone, two-tone (CF + SUP) and, suppressor-tone alone. Each suppressor frequency and intensity combination was repeated 75 times.

The data were obtained from 8 cats: C92101, C92106, C92116, C93030, C93036, C93146, C94009 and C94011. For convenience, they are also referred to as cats A, B, D, E, F, H, I and J, respectively. Cat H's cochlea had a traumatic injury but seemed to recover. That injury was caused by an accidental pulling out of the round window (RW) electrode when the head holder was turned. Although there was no visible damage to the RW membrane under the surgical microscope, the voice-evoked CM responses were much smaller when the RW electrode was re-implanted, and the CAP threshold deteriorated by about 20 dB, compared to comparable responses in other animals and the value observed later in the same animal. The initial AN fiber thresholds were also elevated by about 18 dB. However, after about 24 hours, both the CAP threshold and the fiber thresholds returned to "normal". Thus the data from the first 24 hours of experiment H were excluded from the data analysis. The rest of that animal's data (28 fibers) were included. It should be noted that, except for three fibers with CFs around 3 kHz, all the fibers obtained from experiment H had CFs between 7 and 14 kHz.

The bottom panel of Fig. II-1 shows the post-stimulus time (PST) histogram of one fiber. In the CF-alone segment, there was initially a big response, then an adaptation to a steady state level. Suppression is obvious in the two-tone (CF+SUP) segment. In this particular

example, there is no response to the suppressor tone alone at this intensity level. To analyze the data, we constructed period histograms (64 bins) for the three time windows: CF-alone (75-200 ms), two-tones (250-500 ms) and SUP-alone (575-700 ms), using the period of the suppressor tone as base. The initial portions in each segment were excluded from analysis. The rate vs. suppressor intensity curves were also plotted for the three time windows.

In determining the phase of maximum response to suppressor, the vector phase was used (cf. Chapter 1). The phase of maximum suppression was visually determined from the period histogram due to the non-sinusoidal nature of the period histogram for the two-tone part. In most cases it was quite obvious where the maximum suppression was. When it was not obvious, the mid-point of the points on both sides that have about equal responses was considered to be the point of maximum suppression. In some cases, a secondary suppression phase was also found, often at a higher suppressor intensity, resulting in two minimums in one suppressor period (cf. Fig. II-12). In this case, the phase of the major suppression was used. In an extremely small number of cases, the two suppressions were about equally strong, and those units were excluded from the pool of phase data. The amount by which the maximum suppression *led* the maximum response to the suppressor tone was then termed the *phase lead* of the suppression (cf. Fig. II-4). Each histogram was statistically compared against the uniform distribution, and only those period histograms that have a Rayleigh coefficient less than or equal to 0.001 were included in the phase analysis (Chapter 1).

Analysis of the data included the determination of the synchronization thresholds, rate suppression thresholds and rate excitation thresholds. The lowest suppressor intensity which produced histograms with a Rayleigh coefficient less than or equal 0.001 was considered to be the synchronization threshold. To determine the rate threshold for the responses to

suppressor tone alone, either one of two criteria had to be satisfied. The first criterion was if the SUP-alone rate exceeded by 15% the rate produced by the lowest suppressor intensity used. The second criterion was if the SUP-alone rate exceeded by 15% the rate of the spontaneous activity which occurred in the last 200 ms window that ended the 1-sec repetition period (and thus immediately preceded the next stimulus presentation). The rate threshold to the SUP-alone was then taken as the lowest intensity at which either condition was satisfied.

In determining the rate suppression threshold, we compared the response rates in the two-tone segment and the CF-alone segment. Due to the complication of adaptation in the initial portions of the CF-alone segment, we must consider the time course of adaptation in fibers with different spontaneous rates (SRs) in order to find a realistic criteria. Lower-SR (< 20/s) fibers are known to have longer time courses of adaptation than high-SR (>= 20/s) fibers (e.g. Relkin and Doucet, 1991). In Fig. II-2, the PST histograms of two fibers are shown to illustrate the different time courses of adaptation in low- (<= 1/s) and high-SR fibers. The responses corresponding to the three stimulus conditions are clear: the first segments (0-200 ms) are the responses to CF tone alone, followed by the two-tone responses (200-500 ms, which are suppressed) and the responses to suppressor tone alone (500-700 ms). For the low-SR fiber (lower panel), there is basically no response to the suppressor tone alone. We will concentrate on the CF-alone responses. The bars over the PSTs represent the time windows (75-200 ms) from which the CF-alone rates were estimated. The solid lines below the bars represent the average numbers of spikes in the CF-alone analysis windows. For the high-SR fiber (upper panel), the adaptation process is practically completed before the start of the CF-alone analysis window at 75 ms. But for the low-SR fiber (lower panel), the adaptation process is obviously not complete at the start of the analysis window. Thus the steady-state

response rate for the CF-alone stimulation based on this time window is overestimated for the low-SR fiber. As a result, our program might occasionally report suppression when there was in fact no suppression at all. To avoid this problem, we used a rather strict criterion, requiring the steady-state two-tone rate to be 18% lower than the steady-state CF-alone rate. We understand that this overestimates the suppression threshold for high-SR fibers. The exact percentages used to estimate the suppression threshold (18%) and the rate threshold in the SUP-alone segment (15%) were determined from a sample of about 30 cases by checking the computer reported results against the actual rate curves and PST histograms. Agreement in all 30 cases was satisfactory.

The care and use of animals reported in this study were approved by the Animal Care and Use Committee of the University of Wisconsin-Madison.

Results

1. Phase of suppression and phase lead

Figure II-3 shows typical rate-intensity curves from an AN fiber (fiber D16-4). The frequencies of the CF tone and of the suppressor tone were 1400 and 100 Hz, respectively. As the suppressor intensity was increased, the discharge rate in the two-tone segment gradually decreased. At the same time, the SUP-alone rate increased. At about 85 to 90 dB SPL, the two-tone rate reached its minimum (arrows) and then increased. This non-monotonic two-tone rate-intensity curve is a general characteristic of an AN fiber for "low-side" suppression (Sachs, 1969; Hill and Palmer, 1991; Hill and Geisler, 1992). In Fig. II-4,

Fig. II-4 Period histograms of fiber D16-4. The corresponding rate vs. suppressor-intensity curves are shown in Fig. II-3. The period histograms in all three columns were binned to the suppressor-tone period, so that each abscissa covers the period of the suppressor tone. The phase is always referred to in terms of period (T) of suppressor tone (e.g., 0.45T) in the text. Unless otherwise noted, this convention applies to all figures. The arrows in the two-tone column indicate estimated phase of maximum suppression, while the arrow in the SUP-alone column indicates the "vector phase" of excitation due to the suppressor tone alone. In this and all similar plots, the period histograms in the three (unequally long) time windows were scaled to the two-tone (CF + SUP) case, so that equal discharge rates produced equal bin heights.

the period histograms of the same responses from fiber D16 are shown in three columns corresponding to the three analysis windows. At 75 dB SPL, there is a clear suppression phase in the two-tone histogram (arrow). At 80 dB SPL, the fiber began to show a response to the suppressor tone alone (arrow), with a response peak at phase of about 0.1T (T is the period of the suppressor tone), which lags the phase of suppression at the same intensity (arrow) by about a quarter of a cycle. At 85 dB SPL, there is also a minor suppression phase at about 0.35T. Further increases of suppressor intensity produced more suppression and also larger responses to the suppressor tone alone. At 90 and 95 dB SPL, the two-tone responses reflect mainly the suppressor tone, synchronization to the CF tone being lost.

The suppression phase data for different suppressor frequencies for all of our fibers are plotted in Fig. II-5. For the three lower suppressor frequencies (50, 100 and 200 Hz), the phase of suppression decreased slightly with increasing CF. Comparing suppression phase to the BM phase estimated from the cochlear microphonics (CM) recorded near the round window (horizontal bars on the right-hand ordinate, cf., Chapter 1), it is clear that near the basal region, the suppression phase roughly corresponds to the BM displacement towards SV at 50 Hz, and is between SV displacement and SV velocity at 100 and 200 Hz. For the three higher suppressor frequencies (500, 1000 and 2000 Hz), there is also a trend for the phase to decrease with increasing CF, but due to the considerable scatter, no attempt was made to compute average curves.

Figure II-6 shows the phase by which the major suppression phase leads the (SUP-alone) excitation phase. For a 50-Hz suppressor, the average phase lead is close to a quarter of a cycle in the low-CF region, decreases a little in the CF region around 1 kHz, and then increases to more than a quarter of a cycle at the high-CF end. Similar behavior was seen for

Fig. II-5 Phase of maximum suppression as a function of fiber CF, for different suppressor frequencies. Different symbols encode suppressor intensities as indicated in the figure. More than one point might be entered for one fiber if the phase values at different suppressor intensities are available for the same fiber. The solid lines are the running averages over one octave, in steps of 1/2 octave (outlier points omitted). The small bars on the right hand ordinates indicate estimated phases of BM motion based on CM recorded near the round window (cf., Chapter 1).

Fig. II-6 "Phase-lead" of suppression as a function of fiber CF, for different suppressor frequencies. A "phase lead" is defined as the value by which the phase of maximum suppression leads the vector phase of excitation due to the suppressor tone in the SUP-alone segment. For the three lower-frequency suppressors, the average phase lead is about 1/4T, but for the three lower-frequency suppressors, it is close to 1/2T. For symbols and solid lines see caption of Fig. II-5.

100 and 200 Hz, except that at these two frequencies the minimum phase lead was reached at higher CF regions. But at all three frequencies, the phase-lead tends to be larger at both high and low ends of the CF axis. For higher suppressor frequencies, the phase-lead values are closer to 0.5T. Although there is a lot of scatter in both the phases of suppression (Fig. II-5B-D) and of excitation to the suppressor-alone (cf. Chapter 1), the phase-lead data for the same frequencies are distributed within a much narrower range. This is particularly true for the 1000 and 2000 Hz cases.

Outside points

Although the trends in the lower suppressor frequency data (both the phase of suppression and phase lead) are quite clear, there are some points that are outside the usual ranges (Figs. II-5 and II-6). Some of these outside points are about half a cycle away from the usual range. Similar scattering also exists in the phase of response to the suppressor tone alone (cf., Chapter 1). In Table II-1, we have listed all the fibers in our experiments that showed phase values about half a cycle away from the usual ranges, for any one of the three lower frequencies.

There are several observations to be made. First, fibers showing unusual phase values (either phase of suppression, phase of excitation to suppressor tone alone, or phase lead) usually had CFs either below about 2-3 kHz or above about 9 kHz. Fibers from the mid-CF region rarely show unusual phase values. Most of the high-CF fibers that showed unusual phase values were from experiment H, which had a slight “damage” history but recovered at the time the data were collected. But all 8 experiments contributed to the data presented in Table II-1. The involved fibers (35 total) constitute about 18% of the total number of fibers studied.

Table II-1: Fibers with phases outside usual ranges

		50 Hz			100 Hz			200 Hz		
ID	CF (Hz)	SUP Alone	CF+SUP	PHLEAD	SUP Alone	CF+SUP	PHLEAD	SUP Alone	CF+SUP	PHLEAD
A22	860					OUT		N O R M A L		
A26	1113							OUT	OUT	NML
B2	606		OUT							
B3	393	OUT	OUT	NML	OUT?	OUT?	NML?			
B4	183	OUT?								
B5	900	OUT	OUT	NML				N O R M A L		
B6	850	OUT	OUT	NML	N O R M A L			N O R M A L		
B7	500	OUT	OUT	NML	N O R M A L					
B24	900		OUT		N O R M A L					
D6	13525							NML?	OUT	NML?
D24	1100	N O R M A L			NML	OUT	OUT			
E12	1308				OUT	NML	OUT			
E26	960				N O R M A L			NML	OUT?	NML?
E43	671	N O R M A L						NML	OUT?	OUT
E61	1180				N O R M A L			OUT	NML	OUT
E80	2795	N O R M A L			N O R M A L			OUT	NML	OUT
F17	1600	OUT	NML	OUT	OUT	NML	OUT			
F28	1150				OUT	NML	OUT			
H30	10435								OUT	
H37	9952				OUT	OUT	NML?	OUT?	OUT	NML?
H41	12025							OUT?	OUT	NML?
H42	12820								OUT	
H44	11230				OUT	OUT	NML?	OUT?	OUT	NML?
H58	12765				OUT	OUT	NML?			
H62	12820							OUT	NML	OUT
H66	9830							OUT	NML	OUT
I6	758	OUT?	NML	OUT?	OUT	NML	OUT?			
I7	821	OUT?	NML	OUT?	OUT	NML	OUT?			
I16	12765							OUT	NML	OUT
J4	30000							OUT	OUT	NML
J25	500	OUT	OUT?	OUT						
J40	1220	OUT	NML	OUT	OUT	NML	OUT			
J65	1016					OUT?			OUT	
J77	5054		NML			NML		OUT	NML	OUT
J82	4776		NML			NML		OUT	NML	OUT

Note: PHLEAD = phase lead; OUT = near 180-degrees outside of usual range;
NML = NORMAL = within usual range.

Second, an unusual phase-lead value is mostly caused by an unusual phase of response to the suppressor tone alone (e.g., fibers F17 and H62), only occasionally due to an unusual phase of suppression (e.g., fiber D24 at 100 Hz). Sometimes both the phase of response to the suppressor tone alone and the phase of suppression were about half a cycle away from their usual ranges, but the corresponding phase lead values are within the usual ranges (e.g., fibers B5 and J4). Third, this unusual phase behavior is dependent upon suppressor frequency. That is, a particular fiber may show a phase within the usual range at one frequency and a phase outside the usual range at another frequency, either for the phase of suppression or the phase of responses to the suppressor tone alone (e.g., fibers A22, B5, E61 and E80). Finally, some of the fibers showed a second phase of suppression (see next section). For all the cases that showed an unusual phase-lead value and a second phase of suppression, the phase-lead value falls in the usual range if the second phase is used to calculate the phase lead.

In general, the phase-lead data for higher suppressor frequencies (>= 500 Hz) showed much less variation. But for 500 Hz, there are some points with phase-lead values around zero, which is about half a cycle from the usual range. Figure II-7 shows such an example (D41-9). At 75 dB SPL, this fiber showed a clear suppression at a phase of about 0.38T (arrow). At 85 dB SPL, the fiber responded to the suppressor tone alone, with a peak also at about the same phase (arrow in SUP-alone column). In the two-tone response for this suppressor intensity, although the overall response is reduced, the response near the 0.38T phase actually increased. Further increasing the suppressor intensity to 90 dB SPL resulted in a response reflecting only the suppressor tone. Such a relationship between suppression and excitation (i.e., suppression occurred at the same phase as the response to the suppressor tone when presented alone) was described as a general feature in Sachs and Hubbard (1981, with 500 and 100 Hz suppressors) and Sellick et al. (1982, with 40 Hz suppressor), but it is obviously

not the case in most of our data (Fig. II-6).

Effects of suppressor intensity

Figure II-8 shows the pooled data of the changes of phase of suppression which occurred with increasing suppressor intensity, for several frequencies. Each line represents the phase values obtained from the same fiber at the different suppressor intensities, relative to the phase value occurring at the lowest intensity that showed suppression. A positive value indicates a suppression phase lagging the suppression phase at the first intensity, while a negative value indicates a suppression phase leading the suppression phase at the first intensity. For the 50-Hz suppressor, there is a slight trend upward (more lag) for fibers with CF less than 1 kHz, and downward (more lead) for fibers with CF equal to or greater than 1 kHz (Fig. II-8A and B). This trend is similar to that for the phase of response to the suppressor tone alone at the same suppressor frequency (Chapter 1). The change of phase of suppression did not show any trend at 100 Hz (Fig. II-8C and D) or at other suppressor frequencies. This lack of trends at 100 and 200 Hz is in contrast to similar data for the phases of response to suppressor tone alone, which did show trends. For higher suppressor frequencies, there were sometimes larger phase changes with intensity (e.g., 1000 Hz, Fig. II-8E). In Fig. II-9, we show an example of large phase changes produced by a 1000-Hz suppressor in a high-CF fiber (J94-3). There is no response to the suppressor tone alone up to 80 dB SPL. At the lowest intensity (70 dB SPL), the fiber showed a suppression phase at about 0.25T (arrow). With increasing suppressor intensity, the phase suppression gradually shifted to more lead. At 85 dB SPL, the phase of suppression is at about 0.82T (arrow). However, such large phase changes with intensity are not common in our data.

SR effect

Patuzzi et al. (1984a) observed no difference in the phases of suppression in a pair of high- and low-SR ganglion cells from the basal region of a guinea pig. In Fig. II-10, we plotted the suppression phase data as a function of fiber SR for the 6 suppressor frequencies. We see no obvious trends in the data. In Fig. II-11, the phase-lead data are plotted, also as a function of fiber SR. Due to our use of lower suppressor intensities (<= 95 dB SPL), the fibers with SRs less than about 10/s rarely responded to the three lower-frequency suppressors and thus there are very few points for low-SR fibers in the figure. In the SR region in which fibers responded to the suppressors, there is also no sign of any SR-dependence in the phase-lead.

2. Secondary suppression

It is well documented that in addition to a major suppression which appears at lower suppressor intensities, a second (minor) suppression often occurs when the intensity of the suppressor tone is increased (see Introduction). We also saw such secondary suppressions in our data. For example, fiber D16-4 demonstrated a very small secondary suppression phase at suppressor intensity of 85 dB SPL (Fig. II-4). In Fig. II-12, we show an example (A4) that demonstrated a much clearer secondary suppression. At 75 dB SPL, the lowest suppressor intensity used, suppression is quite obvious; the minimum in the two-tone period histogram is at a phase of about 0.88T. When the suppressor intensity was increased to 80 dB SPL, there is sign of a minor suppression occurring about half a cycle away from the major suppression. This minor suppression becomes obvious at suppressor intensities of 85 and 90 dB SPL (arrows).

Fig. II-10 Phase of suppression as a function of fiber spontaneous rate. The different symbols indicate different suppressor intensities as shown in the figure. The same symbol coding as in Fig. II-5 was used.

Fig. II-11 Phase lead as a function of fiber spontaneous rate. The different symbols indicate different suppressor intensities as shown in the figure. The same symbol coding as in Fig. II-5 was used.

Unlike those reported before (e.g., Sachs and Hubbard, 1981; Sellick et al., 1982; Patuzzi et al., 1984a), not all fibers in our study showed a secondary suppression at higher intensities. In fact, only about one third of the fibers we studied showed secondary suppression. These fibers are listed in Table II-2. The first observation about this table is that secondary suppression existed in all of our experiments. However, there is a higher percentage of fibers (15 out of 28) showing secondary suppressions in experiment H, which contains mainly high-CF fibers. In fact, if we only consider fibers which we used a suppressor with frequency of 200 Hz or less, all except one fiber in that experiment showed secondary suppressions.

The second observation about Table II-2 concerns the dependence of secondary suppression upon suppressor frequency. A fiber did not necessarily show secondary suppressions at all suppressor frequencies studied. For example, fiber D33 showed a secondary suppression at 100 Hz, but not at 50 or 200 Hz. In Fig. II-13, we show the two-tone period histograms of another fiber, A4, for 8 different suppressors (from 100 Hz to 4500 Hz). The period histograms for 200-Hz suppressor of this fiber have been shown in Fig. II-12. Examination of the figure shows that this fiber only exhibited clear secondary suppressions at 200 Hz (arrows), but not at other frequencies, either above or below 200 Hz. The general tendency in the data is for the minor suppression to appear at lower suppressor frequencies (50, 100 and 200 Hz), with 200 Hz being the frequency that largest percentage of fibers showed secondary suppressions. We only saw 5 cases of secondary suppression at suppressor frequency of 500 Hz and we never saw secondary suppressions at 1000 and 2000 Hz, or any frequencies above 2000 Hz that we occasionally used.

The dependence of secondary suppression upon suppressor frequency is also reflected in the thresholds of such suppression. The general trend is that, if the fiber showed secondary

Table II-2: Threshold intensities (dB SPL) at which fibers showed secondary suppressions with suppressors of different frequencies

ID	CF	SR	TH	50Hz	100Hz	200Hz	500Hz	1000Hz	2000Hz
A1	8680	0	8			85		-	-
A2	6040	9.5	-4		85	80	-	-	-
A4	9400	7	-5		-	85	-	-	-
A9	7030	6	0		85			-	-
A18	10000	14	-5			90	85	-	-
A19	9550	21.5	-6			85	-	-	
B12	5880	41.5	11		-	85	-	-	
D16	1300	8	12	90	85	-			
D20	6727	0.5	26	-	95	90	-	-	-
D24	1100	48.5	-3	-	80				
D33	2820	61.5	13	-	90	-			
D43	10240	104	20		95	85	-	-	-
E32	18010	0	33				90	-	
E61	1180	15	14		90	-			
E63	860	17.5	20	95	-				
E71	3415	0	34		90		-		
E80	2795	35.5	17	-	90	-	-		
E82	9130	9.5	9			90	-	-	
F9	18010	103	3				84	-	
F10	16518	38.5	-4				85?	-	
H23	2830	54	26	90	85	80?			
H24	2830	56	23		85	-			
H30	10435	3	19			90	-	-	-
H37	9552	65	0		95	90?	-		
H38	10108	0	15			95		-	-
H42	12820	1.5	9			90		-	
H44	11230	37	7		90	85	-	-	
H47	8275	71	6			85	-	-	
H59	9775	0.5	8		90	90	-	-	-
H62	12820	38.5	-3			90	-	-	-
H66	9830	72.5	-4			85	-	-	-
H68	12020	0	10			90	-	-	-
H70	9620	2.5	5		90		-	-	-
I6	758	21.5	21	95	85				
I7	821	19	-3	90	85				
I12	20000	56.5	23				85	-	-
I14	6040	52	5		90			-	
I15	6040	85.5	8		90	85			
I16	12765	68.5	-1			90	-		

J34	974	40.5	1	85	-			
J40	1220	39	-4	95	80			
J42	1178	56	-4	85	-	-		
J44	400	47	8	85				
J54	4215	102	13	90	85	75	-	-
J55	2815	46	6	90	80	-		
J75	4810	53.5	10	90	85	75		
J77	5054	46	-2	85	80	75	-	-
J80	3020	36	-1		85	-		
J81	3218	25	0	85	80			
J82	4776	33	10	90	85	75	-	-

Note: 1. Dash means only one phase of suppression was observed at all suppressor intensities used.
2. Fibers with data from only one suppressor frequency are not included.
3. Total of 63 fibers showed secondary suppressions at one or more suppressor frequencies, and total of 45 fibers showed peak-splitting (cf. Chapter 1). Seventeen fibers showed both.
4. Also notice that peak-splitting mainly happens in fibers with CF < 2 kHz, while 2nd suppression mainly happens in fibers with CF > 2 kHz.

Fig. II-13 Two-tone period histograms of fiber A4 for stimulation at various intensities at 8 different suppressor frequencies (from 100 Hz to 4500 Hz). The period histograms for 200-Hz of this fiber has been shown in Fig. II-12. This fiber showed clear secondary suppressions *only* at 200 Hz, but not at other frequencies.

suppressions at several suppressor frequencies, the thresholds decreased with increasing frequency. From Table II-2 we can see that no fiber had a higher secondary suppression threshold at 100 Hz than at 50 Hz, and so on. For a particular suppressor frequency, fibers with higher CFs usually have higher thresholds of secondary suppression.

Occurrence of secondary suppression with respect to CF

The data shown in Table II-2 seem to suggest that fibers with higher CFs are more likely to demonstrate secondary suppressions. In Table II-3, we have listed the number of cases in which we saw secondary suppressions for the three lowest-frequency suppressors (50, 100 and 200 Hz), arranged according to fiber CFs. The data from Table II-3 confirm the trend observed in Table II-2. For all three suppressor frequencies, a larger percentage of fibers with higher CFs showed secondary suppressions than those with lower CFs. The CF division seems to be dependent upon the frequency of the suppressor that was used: it was about 1 kHz for a 50 Hz suppressor, 2 kHz for a 100 Hz suppressor and 4 kHz for a 200 Hz suppressor. Moreover, all of the 5 fibers that showed secondary suppressions at 500 Hz had CFs at or above 10 kHz (cf. Table II-2).

Threshold differences between major and minor suppressions

It would be interesting to know the relative thresholds for the major and minor suppressions in those cases that showed two suppressions, since, assuming suppression is due to saturation of OHC receptor currents (Geisler et al., 1990), the difference in thresholds would reflect the degree of asymmetry of the OHC transfer characteristics and/or the variable DC biasing of the inputs. In Fig. II-14, we show the threshold-difference histograms for various groups of

Table II-3 Number of fibers showing secondary suppressions vs. total number of fibers

CF \ SUP(Hz)	50	100	200	all*
<=1K	6 / 35	3 / 34	0 / 6	7 / 53
1-2K	4 / 12	3 / 22	0 / 10	6 / 25
2-4K	4 / 11	12 / 25	2 / 9	14 / 31
4-8K	4 / 5	11 / 14	8 / 9	12 / 19
8-12K		6 / 8	13 / 13	15 / 28
12-16K		1 / 3	4 / 7	5 / 19
>16K			0 / 1	4 / 18

* Fibers showed secondary suppressions at least at one supppressor frequency, including 500 Hz.

Fig. II-14 Number of fibers showing the indicated differences between major and minor suppression "thresholds" (see text). Fibers with lower CFs (<= 2 kHz) mostly had threshold differences of 10 dB. For fibers with CFs between 2 to 4 kHz, the peak in the histogram is shifted to 5 dB. Fibers with higher CFs (> 4 kHz) mostly had threshold differences of zero or 5 dB.

fibers. Fibers with lower CFs (<= 2 kHz) mostly had threshold differences of 10 dB, with only one fiber showing a threshold difference of 0 dB. If we had used higher suppressor intensities, more lower CF fibers would show second suppressions, with larger threshold differences. For fibers with CFs between 2 and 4 kHz, the peak in the histogram shifted to 5 dB. Fibers with higher CFs (> 4 kHz) mostly had threshold differences of zero or 5 dB.

Phase distance between two suppressions

In an attempt to characterize the two suppressions, we measured the phase distance between the two suppressions (i.e., the phase differences between the two minimums in the period histogram), using the convention that the minor suppression *led* the major one. Figure II-15 shows the pooled population data for four suppressor frequencies (50, 100, 200 and 500 Hz). All points are distributed within 0.1 cycle of the line of 0.5T, suggesting that the two suppressions are always about half a cycle apart. Another observation from this figure is that there is a discontinuity near CF of 2 kHz. Since this discontinuity has no counterpart in the phase of suppression vs. CF plot (Fig. II-5), it suggests that fibers near 2 kHz tend not to show secondary suppressions.

We also measured the phase distance between the two peaks in a similar manner, using the convention that the peak just leading the minor suppression *led* the peak just leading major suppression. As can be seen from Fig. II-16, the average distance between the peaks is slightly less than half a cycle.

Degree and phase of secondary suppression

It is common to find fibers showing different degrees of secondary suppressions in all experiments. Occasionally, the major suppression happens at the phase that the minor suppression usually appears. In experiment H, we were able to find several fibers with *similar* CFs that showed different degrees of secondary suppressions. Figure II-17 shows the period histograms of their responses at suppressor intensities of 85 and 90 dB, with a 200-Hz suppressor. Their CFs were between 8-13 kHz and their spontaneous rates ranged from 37 to 71 spikes/second. Their responses to the 200 Hz suppressor tone alone were very small as compared with the two-tone responses, and showed vector phases between 0.7-0.9T. These histograms are arranged, from top to bottom, with increasing degree of suppression in the first half cycle. It is interesting to note that the major suppression happened in the second half cycle for the top two fibers (H62 and H47), while the minor suppression happened in the first half cycle. The suppression in the first half cycle becomes equal in magnitude to the suppression in the second half in fiber H44. In the next fibers, the major and minor suppressions reversed their positions. In fiber H37, the suppression in the first half cycle became the major suppression, while fiber H41 showed only one phase of suppression, and that occurred in the first half cycle.

3. Effect of CF-tone intensity

In addition to varying the intensity of the suppressor tone, we also varied the intensity of the CF tone in some fibers. In Fig. II-18, we show the responses of one fiber (I6) for which the CF intensity was varied. The frequency of the suppressor tone was 50 Hz. This fiber displayed very little response to the suppressor tone alone at the suppressor intensities which we used (column D). When the CF tone was presented at 35 dB SPL (column A), the fiber began to show a phase-specific suppression at a suppressor intensity of 75 dB SPL (arrow).

Fig. II-17 Different degrees and directions of secondary suppressions in neighboring fibers. Unlike other figures, the responses at different suppressor intensities are arranged by column. Each row is for a different unit. All five fibers were from the same experiment (C93146) and the same 200-Hz suppressor was used. From top to bottom: fiber H62-2, CF = 12820 Hz, SR = 38.5/s, TH = -3 dB SPL, SUP-alone response phase 0.757T; fiber H47-8, CF = 8275 Hz, SR = 71.0/s, TH = 6 dB SPL, SUP-alone response phase 0.937T; fiber H44-8, CF = 11230 Hz, SR = 37.0/s, TH = 7 dB SPL, SUP-alone response phase 0.827T; fiber H37-9, CF = 9552 Hz, SR = 65.0/s, TH = 0 dB SPL, SUP-alone response phase 0.803T; fiber H41-5, CF = 12025 Hz, SR = 55.5/s, TH = -2 dB SPL, SUP-alone response phase 0.736T. Notice that the three pivotal ones (H62-2, H44-8 and H41-5) had very similar CFs.

Fig. II-18 The two-tone responses of one fiber at different CF and SUP intensities (C94009, fiber I6, CF = 758 Hz, SR = 21.5/s, TH = 21 dB SPL). The frequency of the suppressor tone was 50 Hz. When the CF tone (800 Hz) was presented at 35 dB SPL (column A), the fiber began to show a phase-specific suppression at a suppressor intensity of 75 dB SPL (arrow). At 95 dB SPL, there is also a strong secondary suppression at phase of about 0.5T (arrow), the depth of which decreased with increasing CF intensity. The threshold of suppression increased with CF intensity (columns B and C) but the suppression phases stayed approximately constant. See text for details.

As the suppressor intensity was increased, suppression became more obvious. At 95 dB SPL, there is also a strong secondary suppression at phase of about 0.05T (smaller arrow).

The two-tone responses of this fiber with CF intensity of 40 dB SPL are shown in column B. As expected, the overall responses at all suppressor intensities were increased, as compared to the responses when the CF tone was presented at 35 dB SPL (column A). Several additional observations can be made. First, there was no suppression when the suppressor was presented at 75 dB SPL, the intensity at which suppression was first seen when CF tone was presented at 35 dB SPL (column A). It required a suppressor intensity of 80 dB SPL to produce a phase-specific suppression (arrow). At a suppressor intensity of 95 dB SPL, there is also a secondary suppression (smaller arrow), but it is shallower than the one in column A. The final observation is that the phases of suppressions did not change with increasing CF intensity: the major suppression was still at a phase of about 0.5T and the minor suppression at a phase of about 0.05T.

Column C of Fig. II-18 shows the same fiber's two-tone responses with the CF tone presented at a still higher intensity, 50 dB SPL. Although the intensity of the CF tone was 10 dB greater than that used for column B, the responses did not change very much, indicating that the responses saturated. The minor suppression at suppressor intensity of 95 dB SPL (smaller arrow) was somewhat shallower than those in both column A and column B, but the phases of suppressions did not change. Comparing the responses shown in column A and column C, although there was a 15-dB increase in CF intensity, the threshold of suppression only increased by about 5 dB. This relative insensitivity to CF intensity is a common feature in our data.

Figure II-19 shows the responses from the same fiber as in the previous figure, but with a suppressor frequency of 100 Hz. The responses in this figure bear some common features to those shown for 50-Hz suppressor: the phases of suppressions did not change with CF intensity, and the suppression threshold increased only slightly as CF intensity was increased. Of particular interest are the responses at higher suppressor intensities. In column A (CF intensity of 30 dB SPL), there are responses between the peaks at suppressor intensity of 85 dB SPL (bar), which must be due to drive from the suppressor tone (cf., column E, bar). But in column D (CF intensity of 50 dB SPL), there are no comparable response components (bar). Even more striking differences existed in the responses for a suppressor intensity of 90 dB SPL (bottom row). These data show clearly that complex interactions can occur between the CF tone and the suppressor tone: in addition to the suppressive effect of the suppressor tone upon the CF tone, the CF tone also can affect the fiber's responses to the suppressor tone.

Similar types of complex interactions are shown for another fiber (J55) in Fig. II-20. The basic response characteristics are similar to those shown in Figs. II-18 and II-19. With increasing CF intensity, the threshold of suppression was slightly increased. When the CF tone was presented at 13 dB SPL (column A), a clear suppression was seen when suppressor intensity was 75 dB SPL (arrow); but when the CF intensity was 28 dB (column D), the suppression first appeared at a suppressor intensity of 80 dB SPL (arrow). The phases of neither the major nor minor suppressions changed with CF intensity. The complex interactions occur at a suppressor intensity of 90 dB SPL (bottom row). This fiber responded to the suppressor tone alone (column E) with appreciable probability over the 0.7T to 0.9T region (bar), the region in the major suppression in the two-tone response. At a CF intensity of 13 dB SPL (column A), the two-tone response was very similar to that

Fig. II-19 Two-tone responses from the same fiber as in Fig. II-18, at different CF and SUP intensities. The suppressor frequency was 100 Hz. Of particular interest is the responses at higher suppressor intensities. In column A (CF intensity of 30 dB SPL), there are responses between the peaks at suppressor intensity of 85 dB SPL (bar), which must be due to excitation by the suppressor tone (see responses to the suppressor-tone alone in column E, bar). But in column D (CF intensity of 50 dB SPL), there are no similar responses between the small peaks (bar). Similar phenomenon also existed in the responses when suppressor intensity was 90 dB SPL.

Fig. II-20 The two-tone responses of single fiber at different CF and SUP intensities (C94011, fiber J55, CF = 2815 Hz, SR = 46.0/s, TH = 6 dB SPL). The suppressor frequency was 100 Hz. At a CF intensity of 13 dB SPL (column A) and a suppressor intensity of 90 dB SPL, the two-tone response was very similar to the suppressor alone response at the same suppressor intensity (column E). With increasing CF intensity, the responses in the second half cycle gradually decreased for the 90-dB suppressor case, especially at a phase of 0.7-0.8T (bars). Arrows indicate estimated onset of suppression.

evoked by the suppressor alone (column E), showing the same gradual response buildup in the 0.7T to 0.9T region (bar). However, with increasing CF intensity, the responses in this region gradually decreased, almost disappearing at the highest CF intensity (column D, bar). These data show that the responsiveness to the suppressor tone was suppressed by the CF tone in that region.

4. Synchronization threshold vs. rate suppression threshold

Javel et al. (1983) studied the low-side suppression in auditory-nerve fibers of cats and found that the suppression threshold was *always* higher than the synchronization threshold. For comparison purposes, we chose data only from fibers with SRs of 10/s or greater. Figure II-21 shows the rate suppression threshold as a function of the synchronization threshold in the SUP-alone segment. The diagonal lines indicate equal rate suppression thresholds and synchronization thresholds. With lower frequency suppressors (50, 100 and 200 Hz, Fig. II-21A), both thresholds were generally higher than those with higher frequency suppressors (500, 1000 and 2000 Hz, Fig. II-21B). But for all suppressor frequencies, the data seem to be scattered on both sides of the diagonal line, with slightly more points falling above the line (cf., figure caption).

Figure II-22 shows the rate suppression threshold as a function of the rate threshold of the response in the SUP-alone segment. In this figure, almost all of the data points lie below the diagonal line, indicating that the rate threshold was almost always higher (more intense) than the suppression threshold. Of particular significance is that this relationship holds not only for higher-frequency suppressors (panel B), but for lower-frequency suppressors as well (panel A), since it was believed that very low frequency, non-excitatory suppressors could

not cause appreciable rate suppression (cf., Cheatham and Dallos, 1992a, 1995). Our data clearly indicate otherwise. The data shown in Fig. II-18 (fiber I6, low CF, suppressor frequency = 50 Hz) are a clear example. In Fig. II-23, we show another example from a higher-CF fiber (J56-4) in which a 100-Hz suppressor was used. Even at a suppressor intensity of 80 dB SPL, there was significant suppression, but there was no response to the suppressor alone until the suppressor intensity was 95 dB SPL.

In Fig. II-24, the threshold data shown in Figs. II-21 and II-22 are plotted for groups of fibers having different CFs. We also added an additional measure to these graphs, the threshold of synchronization to the suppressor tone in the *two-tone* segment. Two major observations can be made. First, regardless of fiber CF and the frequency of the suppressor, the synchronization threshold in the two-tone segment is always the lowest among the four measures, and the SUP-alone rate threshold is always the highest. Second, the threshold of synchronization (in the SUP-alone segment) and the threshold of rate suppression are generally very close to each other, but for lower frequency suppressors and in fibers with lower CFs (< 4 kHz), the synchronization threshold is usually below suppression threshold, a trend which is statistically significant ($p < 0.01$ and 0.001, F-test).

5. Complete suppression

In some low-SR fibers, the response due to the CF tone can be almost or completely suppressed as previously described by others (e.g., Fahey and Allen, 1985). A similar phenomenon was also observed in this study. Ideally, the term *complete suppression* means that there is no response at any phase in the period histogram, either due to CF tone or to the suppressor tone. In practice, this condition is difficult to meet, so instead, if the remaining

Fig. II-24 The average behavior of the data shown in Figs. II-21 and II-22 is plotted for fibers grouped into four CF categories. We also added an additional measure, the threshold of synchronization to the suppressor tone in the *two-tone* segment.

response is reasonably small (e.g., less than 2% of the response in CF-alone segment, in terms of absolute rate), we consider it to be complete suppression.

Figure II-25 shows the responses of a low-SR fiber (J12), which showed no response to the suppressor tones alone even at 95 dB SPL (columns B and D, with suppressors of 50 and 100 Hz respectively). With a 50-Hz suppressor, there was a strong suppression at phase 0.55T (column A) at a suppressor intensity of 80 dB SPL. As the suppressor intensity was increased, more and more responsiveness was suppressed. At 90 dB SPL, there was a secondary suppression at phase of about 0.05T. At 95 dB SPL, there was basically no response in the two-tone histogram, except for the two small peaks at phases of about 0.2T and 0.85T respectively (arrows). The same fiber showed complete suppression at 90 dB SPL with a 100-Hz suppressor (column C, arrow).

The responses of another low-SR fiber (A9) to higher frequency suppressors (1000 and 1750 Hz) are shown in Fig. II-26. With a 1000-Hz suppressor, the fiber showed significant suppression at 65 dB SPL and there was very little remaining response at 70 dB SPL (column A), no more than to the suppressor tone alone (column B). With a 1750-Hz suppressor, the suppression threshold was lower than that in the 1000 Hz case. At 55 dB SPL, there was already obvious suppression (column C). At 65 dB SPL, the two-tone rate was very low, about the same as to the suppressor tone alone. A further increase of suppressor intensity resulted in response increases, apparently to the suppressor tone only (columns C and D). Thus, although the fiber showed severe suppression, it never reached complete suppression at this suppressor frequency, due to the responses to the suppressor tone.

There were total of 18 fibers (out of ~190) which showed complete suppression at one or

Fig. II-25 Examples of complete suppression (C94011, fiber J12, CF = 1138 Hz, SR = 0.5/s, TH = 11 dB SPL). The suppressor frequency was 50 Hz in columns A and B and 100 Hz in columns C and D. The CF tone was presented at 28 dB SPL. With a 50-Hz suppressor, there was basically no response in the two-tone histogram at 95 dB SPL, except for the two small peaks at phases of about 0.2T and 0.85T respectively (arrows). The same fiber showed complete suppression at 90 dB SPL with a 100-Hz suppressor (column D, arrow).

Fig. II-26 Examples of nearly complete suppression (C92101, fiber A9, CF = 7030 Hz, SR = 6.0/s, TH = 0 dB SPL). The CF tone was presented at 10 dB SPL. The suppressor frequencies were high (1000 and 1750 Hz), compared to those in Fig. II-25. With a 1000-Hz suppressor, the fiber showed significant suppression at 65 dB SPL and there was practically complete suppression with very little remaining response at 70 dB SPL. With a 1750-Hz suppressor, the suppression threshold was lower than that in the 1000 Hz case. However, the fiber begin to respond to the suppressor tone alone at about 60 dB SPL. Thus, although the fiber showed severe suppression, it never reached complete suppression at this suppressor frequency, due to the excitation caused by the suppressor tone.

more suppressor frequencies, most of which involved 500 or 1000 Hz suppressors. It was more difficult to get complete suppression with either lower or higher frequency suppressors. In only one case did we see a nearly complete suppression using a 50-Hz suppressor. For low-frequency suppressors, the CF component was often greatly attenuated but not completely suppressed (cf. Fig. II-25). But for higher suppressor frequencies, often there were appreciable responses to the suppressor (cf. Fig. II-26).

By our definition of complete suppression, there should be no high-SR fibers showing complete suppression, due to their spontaneous firings. However, the CF-driven responses can be "suppressed" in some fibers, through "synchrony suppression" mechanism (Javel et al., 1983). In Fig. II-27, we show an example (E47-6) of this type which demonstrates temporally how the synchronization is shifted from CF tone to the suppressor tone without significant reduction of response rate. This fiber had a low CF so the responses to the CF-tone were phase-locked. To help visualize the phase relationship, a vertical line was drawn through the second response peak in the two-tone histograms (column A) and a similar line was drawn in the histograms of suppressor alone (column B), at exactly the same phase. At 80 dB SPL, the second peak becomes the major remaining response peak in the two-tone histogram. At this intensity, there is also a significant response to the suppressor tone alone. However, at 85 dB SPL, the response peaks in both columns are exactly in phase, suggesting that the response due to the CF tone was completely "suppressed". Notice that there was very little change in response rates between these two intensities.

6. Some details in the temporal response: Possible role of IHCs

The input to the IHC in the two-tone case can be viewed as the sum of the suppressor tone

Fig. II-27 Examples of complete suppression of the CF component from a high-SR fiber (C93030, fiber E47-6, CF = 785 Hz, SR = 28.0/s, TH = 24 dB SPL). The CF tone was presented at 35 dB SPL. To help visualize the phase relationships, a vertical line was drawn through the second response peak in the two-tone histograms (column A) and an identical line was drawn in the histograms of suppressor alone (column B). At 80 dB SPL, the second peak becomes the major remaining response peak in the two-tone histogram. At this intensity, there is also a significant response to the suppressor tone alone (column B). However, at 85 dB SPL, the response peaks in both columns are exactly in phase. Notice that there was basically no change in response rates at these two intensities.

and the CF tone. Thus, at the IHC input, the CF tone is presumably riding on the suppressor tone. If the combined input in the two-tone case is large enough, it may push the cell's "operating point" to the saturating region of the IHC transfer function (Russell et al., 1986), thus "suppressing" the CF component.

Figure II-28 shows an example (fiber I12) in which a 1000-Hz suppressor tone was used. At 75 dB SPL, this fiber showed some suppression (cf. dashed lines and arrow), which is not phase specific. To emphasize that this is not due to our choice of analysis windows, the PST at this intensity is shown in the insert, where there is obvious rate suppression between 200 and 500 ms, the two-tone segment. At a suppressor intensity of 80 dB SPL, a phase specific suppression occurred in the two-tone period histogram, with a minimum at a phase of 0.5-0.6T (arrow). Although the two-tone period histogram resembles its counterpart in the SUP-alone column at the same intensity, the phases of the minimums in the two histograms do not match exactly. Further increasing the suppressor intensity to 85 dB SPL produced period histograms in the two-tone and SUP-alone segments almost exactly alike in phase and shapes, which is the characteristic of suppression produced by the IHC stage (Chapter 4).

For high CF fibers, it is difficult to know whether the response in the two-tone segment at higher suppressor intensities is due to the CF tone or to the suppressor tone, since there is no synchrony information produced by the CF tone. In the next two figures, we show the responses from two low CF fibers. In the example shown in Fig. II-29 (fiber E28-5), a suppressor of 100 Hz was used. At a suppressor intensity of 75 dB SPL, this fiber showed some suppression, with a minimum at a phase of about 0.71T. At 95 dB SPL, the response in the two-tone segment is basically the same as the response to the suppressor tone alone. Now we focus on the two-tone responses at 85 and 90 dB SPL. At 85 dB SPL, the second

response peak (at a phase of 0.1T, arrow) is just slightly shorter than the third one, and is much taller than the first one. At 90 dB SPL, this second peak is greatly reduced (arrow), while the first and third peaks, although also reduced, are much taller. At the same intensity, there is also a notch in the SUP-alone response, which coincides with the phase of the second peak (arrow).

Another low-CF example (D24-4) is shown in Fig. II-30. The suppressor frequency was also 100 Hz. This fiber responded to the suppressor tone alone even at 70 dB SPL, the lowest suppressor intensity which we used. At 75 dB SPL, there is suppression in the two-tone response, with the minimum occurring at about 0.4T (arrow). At 80 dB SPL, the suppression at this same phase becomes deeper. The highest response peak in the two-tone part is now taller than the response peaks in the CF-alone part. Also, there is another suppression at a phase of 0.8T, which is leading the phase of the response peak (0.1T) in the SUP-alone column by about 0.3 cycle. At 90 dB SPL, although the shape of the two-tone period histogram is identical to that of the SUP-alone one, the synchrony to the CF tone was still preserved. At all intensities, the suppression phases near 0.4T coincide with the minimums in the SUP-alone response. Thus, in these two low-CF examples, the two-tone responses are also synchronized to the suppressor tone.

Discussion

1. Summary of results

The responses of AN fibers to a CF tone and a low-side suppressor tone were studied in cats.

The main findings can be summarized as follows.

a. The response of a fiber to the CF tone was suppressed by a low-side suppressor tone, in a phase-specific manner. The phase of maximum suppression within the suppressor tone period is a monotonic function of fiber CF. On average, the phase of suppression led the phase of response to the suppressor tone (so called "phase lead") by about a quarter of a cycle for lower-frequency suppressors (50, 100 and 200 Hz), and was close to half a cycle for higher-frequency suppressors (500, 1000 and 2000 Hz). For lower frequency suppressors, the phase lead is also CF-dependent, with a bigger average value at both very low and very high CFs. Both the phase of suppression and the phase of response to the suppressor tone alone are independent of fiber SR.

b. Some fibers also show a secondary (minor) suppression at higher suppressor intensities, which is always about half a cycle away from the first (major) one. Fibers with higher CFs (> 2 kHz) are more likely to show a secondary suppression than those with lower CFs. The differences between threshold intensities for the major and minor suppressions are CF-dependent: lower CF fibers usually show differences of 10 dB or greater, while higher CF fibers show smaller differences. The occurrence of secondary suppression is dependent upon suppressor frequency, that is, a fiber may show a secondary suppression at only one suppressor frequency but not at any others. The secondary suppressions are usually restricted to lower-frequency suppressors (<= 200 Hz). We did not see a secondary suppression with a suppressor frequency of 1000 Hz or greater.

c. The phases of maximum suppressions (both the major and minor ones) are not affected by the intensity of the CF tone, but the threshold of suppression is increased slightly when the

CF intensity is increased. At higher CF intensities, the CF tone can suppress the response to the suppressor tone at some specific phases.

d. Non-excitatory low-frequency suppressors can also give rise to significant suppression. For any one fiber (with SR > 10 /s), the threshold of synchronization to the suppressor tone in the two-tone segment was usually the lowest (most sensitive), while the SUP-alone rate threshold was highest. The threshold of suppressor synchronization in the SUP-alone segment and the threshold of rate suppression fell somewhere in between.

e. In some low-SR fibers, complete suppression can be seen. In general, it is more difficult to achieve complete suppression with either low- (< = 100 Hz) or high-frequency (>= 2000 Hz) suppressors.

2. Comparison with previous two-tone suppression studies

Phase of suppression

There are very few studies of the temporal aspects of two-tone suppression in the AN-fiber literature (Romahn and Boerger, 1978; Sachs and Hubbard, 1981; Sellick et al., 1982; Patuzzi et al., 1984a). In the Romahn-Boerger study, a 50-Hz tone was used to provide biasing for a short train of tone bursts at CF placed at different phases of the 50-Hz tone. They found that in only 7% of the fibers were the discharges due to the CF tone burst affected by the biasing tone. This result is obviously at variance with other studies and also with this study. Sachs and Hubbard (1981) studied the two-tone responses of AN fibers of cats, using low-frequency suppressors (<= 1 kHz). Most of the fibers they studied had CFs

above 5 kHz. For 500 and 100 Hz suppressors, there are usually two phases of suppression: a major one at lower intensities, and also a minor one at higher intensities. They concluded that at low suppressor intensities the phase of the suppressor tone which causes the instantaneous rate to increase when presented alone causes a rate decrease when added to a CF tone. This corresponds to a zero phase-lead in our case. Such zero phase-lead cases also exist in our data, but only in small numbers (Fig. II-6B and D, Fig. II-7). Since Sachs and Hubbard (1981) did not show population data, it is difficult to compare the phases from their study directly with ours, particularly since the data which they published display considerable scatter. For example, in one of their low-CF examples (1500 Hz), the fiber showed a phase lead of 1/4 cycle with 50-Hz suppressor (their Fig. 9), which agrees with the major trend in our comparable data (e.g. Fig, 6A).

Patuzzi, Sellick and colleagues (Sellick et al., 1982; Patuzzi et al., 1984a) recorded from the ganglion cells of guinea pigs, in the high-CF region (> 10 kHz), while using a 40-Hz suppressor tone. They observed suppression patterns similar to those of Sachs and Hubbard (1981), a major suppression which appeared at lower intensities and a minor one at higher intensities. At lower intensities, the two-tone response was enhanced at one phase and suppressed at another. There was no example in their papers in which the SUP-alone excitation phase and the two-tone suppression phase were collected at the same intensities. If the SUP-alone excitation phases at higher intensities are used, the two examples they showed (Figs. 4 and 5 of Sellick et al., 1982) would give phase lead values of about 1/4 and -1/4 cycles respectively. The -1/4 cycle phase lead did not agree with our 50 Hz data (Fig. II-6A). Although we have some examples of -1/4 cycle phase lead in our 50-Hz data for low-frequencies fibers (Fig. II-6A), their 1/4 cycle phase-lead example is much more in keeping with what we observed. Again, lack of population data prevents further comparison.

Although different suppressor frequencies were used, there is a common feature in the studies of both Sachs and Hubbard (1981) and Sellick et al. (1982). Both groups concluded that zero phase-lead was a general finding and presented an example to support their conclusion. However, both groups also presented data with quite different phase relationships. Based on a more extensive sample, we believe our data to be more general.

Two-phased suppression pattern

In studies by Sachs and Hubbard (1981) and by Sellick, Patuzzi and colleagues, the two-phase suppression pattern was a general characteristic. In our study, we did not see this pattern in all fibers. Instead, we found a CF dependence of the occurrence of the secondary suppression: a large percentage of high-CF fibers (> 2 kHz) showed secondary suppressions while only a small number of low-CF fibers did. Considering the fact that in previous studies data were collected only or mainly from high-CF regions, and that higher suppressor intensities were used (which probably would cause more secondary suppressions), our results are probably not significantly different from previous studies in this regard. For the fibers with higher CFs, we only found limited cases of secondary suppressions with 500-Hz suppressor in our data, while in Sachs and Hubbard (1981) the secondary suppression seems to be a general characteristic with that suppressor frequency. However, in their 500-Hz examples, the fibers were responding strongly to the suppressor tone alone (their Fig. 2). Thus, the pattern they saw might be due to a combination of CF suppression and the response to the suppressor tone alone, rather than being a pure suppression pattern. They “rarely” saw secondary suppressions with a 1000-Hz suppressor, which agrees with our results.

Effect of CF intensity

Sachs and Hubbard (1981) have studied the effect of CF intensity on the phase of suppression. In their 100-Hz suppressor example (their Fig. 8), the response to the suppressor alone (presented at 94 dB SPL) was very small and the CF intensity did not affect the phase of suppression. This result agrees with our findings (Figs. II-21 to II-24) and those of Patuzzi et al. (1984a). However, in most other examples they showed (their Figs. 3-6), they used higher frequency suppressors (500 or 1000 Hz). In those cases, the fibers were responding to the suppressor tone alone more vigorously. At lower CF intensities, the CF response might be completely suppressed (e.g. their Fig. 3) so that the two-tone response was very similar to the SUP-alone response. At higher CF intensities, the phases of suppressions did not change with CF intensity, which agrees with our data and their own example with a 100 Hz suppressor. In those examples obtained with higher frequency suppressors, their data also showed phase-specific reductions of the suppressor-tone components by the CF tone in a manner which is similar to ours (Fig. II-20).

Suppression patterns on BM

There have been several studies of 2TS in the basal region of BM (Patuzzi et al., 1984b; Ruggero et al., 1992; Rhode & Cooper, 1993; Nuttall and Dolan, 1993). All except that of Nuttall and Dolan (1993), which used suppressors with frequencies close to CF, would allow us to compare the suppression patterns with those observed on AN. Patuzzi et al. (1984b) studied the two-tone responses of the guinea pig's BM with a low-frequency suppressor tone (33 Hz). Although their relatively insensitive Mössbauer method prevented them from obtaining the fine temporal details of the waveforms, they were able to show two phases of suppression in one cycle of the suppressor tone: a major one which corresponded to maximum displacement of the BM towards ST and a minor one corresponding to maximum

SV displacement which was observed at higher suppressor intensities. Using a velocity sensitive laser interferometer, Ruggero et al. (1992) studied the chinchilla BM responses, with higher suppressor frequencies (300-500 Hz). They also found a similar two-phased suppression pattern in their preparation, but the two suppression phases corresponded to maximum BM velocity towards ST and SV, respectively. Recently, Rhode and Cooper (1993) used a displacement sensitive laser interferometer to study the BM response of the hook region of the cat. In one of their examples, they used a 1000-Hz suppressor and a 34-kHz CF tone. The basic response pattern was similar to that of the previous two studies, i.e., a major suppression at lower suppressor intensities and also a minor one at higher suppressor intensities. The major suppression roughly corresponded to BM displacement towards ST. In all three studies, and particularly the later two which used the much more sensitive laser interferometer, the temporal distance between the two suppression phases was about half a cycle.

These temporal suppression patterns agree very well with our findings in higher CF fibers (e.g., Fig. II-12) and those of the other studies on AN fibers (Sachs and Hubbard, 1981; Sellick et al., 1982; Patuzzi et al., 1984a). The distance between the two suppression phases in our study was also about half a cycle away (Fig. II-15). But such two-dip suppression patterns were usually observed for suppressor frequencies of 200 Hz or less. We only found limited examples with a 500-Hz suppressor and none with 1 and 2-kHz suppressors.

Relationships among synchrony, excitation and suppression

Javel et al. (1983) studied the two-tone synchrony suppression in the AN fibers of cats, using changes in discharge synchronization as an indicator. According to them, such a

synchronization-based measure would enable a greater number of potential suppressor tones to be studied, particularly in the cases where the suppressor tone itself is excitatory. In our study, we used the traditional discharge rate measure for suppression. Since we only included data from fibers with SR of 10/s or higher in our summary figures (Figs. II-21, II-22 and II-24), those data would be roughly comparable to theirs. In Figs. II-21 and II-22, we plotted the suppression threshold as a function of discharge rate threshold and of synchronization threshold respectively, similar to plots Figs. 7a and 8a of Javel et al. (1983). The small difference between their Fig. 7a and our Fig. II-22 (plotting suppression threshold as a function of discharge rate threshold) probably can be explained by our method of obtaining the rate suppression threshold (see Methods section), which overestimated the suppression threshold for fibers with higher SRs. However, the difference between their Fig. 8a and our Fig. II-21 (plotting suppression threshold as a function of synchronization threshold) can hardly be accounted for. In their case, a fiber's suppression threshold is *always* higher than synchronization threshold, while in our case, the averages are almost equal. Our particular criteria of rate suppression (which tends to overestimate the suppression threshold) would serve to *increase* the difference between our two studies. This difference did not seem to be accounted for by the data samples either. In our case, we only studied those fibers which showed some rate suppression with a short two-tone paradigm. However, unless Javel et al. (1983) did not study fibers which showed clear rate suppression (highly unlikely), they should at least have data points that overlap with our ranges. Considering the sharp contrast in their and our data which came from the same species, we can offer no reasonable explanation here.

3. Mechanisms of two-tone suppression

BM mechanism

The two-tone suppression seen at the neural level has been attributed to its counterpart at the BM (Patuzzi et al., 1984b; Ruggero et al., 1992; Rhode and Cooper, 1993; Cheatham and Dallos, 1995). Our findings in this study seem to agree with this view. For example, the temporal patterns of AN discharges we observed were similar to those observed on BM. At both BM and AN level, there can be two suppression phases in one cycle of the suppressor tone, a major one at lower intensities, and also a minor one occurring at higher intensities. The distance between the two suppression phases is about half a cycle for both AN (Fig. II-15) and BM (Ruggero et al., 1982; Rhode and Cooper, 1993). The fact that the suppression pattern can be predicted by a model using the OHC saturation characteristics (Chapter 4) also supports the idea of BM suppression. Finally, this view is also consistent with the fact that only the responses to tones placed in the tip region of the tuning curve (i.e., the part boosted by the active process) can be suppressed.

Role of IHC and later stages

However, there are several things which are not accounted for by the BM theory. First, on AN fibers we can see non-excitatory suppressors suppressing the CF response (e.g., Fig. II-23), but on BM there has been no report of non-excitatory, low-side suppressors producing suppression. In fact, when suppression of the CF component occurs, the total response at the BM level is always greater than the CF-alone response with a low-side suppressor is used, but with a high-side suppressor the total response can be lower than the CF-alone response

(Rhode and Cooper, 1993; Cooper, 1995). Second, for higher frequency suppressors, we did not see two-phased suppression patterns on the AN, such two-phased suppression patterns were seen at the BM level (Rhode and Cooper, 1993). The phase relationship between suppression and the excitation to the suppressor tone was also different for these high-CF fibers when higher frequency suppressors were used: on the BM level the phase lead is about a quarter of a cycle (Rhode and Cooper, 1993), but on the neural level it is close to half a cycle (Fig. II-6). Thirdly, there is a quantitative difference between the two-tone response in high- and low-SR fiber (Chapter 3), and in some low-SR fibers, we saw complete suppression (Fig. II-25), which was never observed at or before IHC stage. Finally, our computer modeling suggested that the suppression in the data shown in Figs. II-28 to II-30 was at least partly due to the IHC stage (Chapter 4). All the above suggested roles for the later stages leading to spike generation on the AN fiber, predominately the IHC. Such roles will be further explored in Chapters 3 and 4.

Difference between basal and apical responses

There has been suggestions that the apical and basal regions of the cochlea may be working differently (e.g., Cooper and Rhode, 1995). In this study, we also found some differences between the two-tone responses of fibers from the apical and basal regions. In particular, fibers with higher CFs tend to show more secondary suppressions (Table II-3) and the threshold differences between the major and minor suppressions are usually smaller, suggesting that the transfer characteristics of OHCs from the basal region are more symmetric (cf., Chapter 4). This in fact agrees with the in vivo recordings from single OHCs (e.g., Russell et al., 1986; Dallos, 1986). The fact that no fibers with CFs near 2 kHz showed secondary suppressions (Fig. II-15) suggests a division around 2 kHz. The differential rate of

growth of suppression for both low-side and high-side suppressions for basal and apical fibers also seem to have a division line near 2 kHz (Delgutte, 1990). Differences in the slopes of AN-fiber spatial tuning curves also suggest a division in character near this same characteristic frequency (Geisler and Cai, 1995). Thus, there is growing evidence that the basal and the apical cochlea are working in different ways, at least quantitatively.

Variation in the phase data

Although both the phase of suppression and phase lead data showed clear trends, there is some scattering in the data points (Figs. II-14 and II-15). As summarized in Table II-1, unusual phase-lead values are mostly caused by unusual phases of responses to the suppressor tone alone; only in a small number of cases are they caused by unusual phases of suppression. In some other cases, both the phase of suppression and the phase of response to suppressor tone are outside their usual ranges, but their effects cancel each other, giving a phase-lead value in the usual range. Thus it seems that in most cases the determinants of the phase of suppression and phase of response to the suppressor tone are separated. This view is supported by the fact that only a small percentage of fibers showing either peak-splitting (cf. Chapter 1) or secondary suppression showed both types of nonlinearity, and by the examples that fibers with similar CFs from the same experiment responded to the 200-Hz suppressor tone alone at similar phases but showed dramatically different suppression patterns (Fig. II-17).

The unusual phase of suppression seems to be accounted for mainly by the direction of the OHC asymmetry (Chapter 4). The unusual phases of responses to the suppressor tones, however, are probably not related to the OHC function or BM macromechanics, since they

do not seem to be strongly related to the unusual phase of suppression. Instead, the micromechanics or the IHC stage might be responsible. If this is indeed the case, it would suggest some degree of separation between BM micromechanics and the later stages, i.e., the factors affecting BM macromechanics might not affect the micromechanics or the IHC stage to a large degree, and vice versa.

Dependence on suppressor frequency

There are several aspects of suppression that are dependent upon the suppressor frequency. One of them is the phase of suppression and phase lead. For some fibers (cf. Table II-1), the phase of suppression or the phase lead is outside the normal range at one frequency but not at others. The phase-lead is generally around a quarter of a cycle for suppressor frequencies of 200 Hz or less, but is close to half-a-cycle for suppressor frequencies at or above 500 Hz. For suppressors of 200 Hz or less, the phase lead is also CF-dependent: at both high and low CF ends, the phase lead tends to be bigger. But the CF divisions were dependent upon the suppressor frequency. Another major aspect is the secondary suppression, which does not appear to happen at all suppressor frequencies (Table II-2). In particular, they never occurred in our 1000 and 2000-Hz data. The frequency division again seems to be around 500 Hz. For lower suppressor frequencies, the CF above which more fibers show secondary suppressions are 1, 2 and 4 kHz for 50, 100 and 200 Hz suppressors, respectively (Table II-3). Such CF divisions are similar to the suppressor frequency dependence of the direction of the change of phase of SUP-alone response with intensity (Chapter 1).