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Late auditory evoked potentials in elderly long-term hearing-aid users with unilateral or bilateral fittings

Second revision

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1 **Abstract**

2 This study investigated the effects of long-term unilateral and bilateral amplification on
3 central auditory processing in elderly people with symmetrical hearing loss using late
4 auditory evoked potentials. It was hypothesized that in the unilateral setting stimulation
5 of the aided ear would yield an acclimatization effect with larger amplitudes and shorter
6 latencies of the components P1, N1 and P2 compared to those of the unaided ear.
7 Auditory evoked potentials were elicited by 500, 1000 and 2000 Hz pure tones at 55,
8 70 and 85 dB SPL presentation level delivered either to the left or right ear. Unilaterally
9 and bilaterally fitted experienced hearing-aid users and a control group of normally
10 hearing adults, all aged at least 60 years, participated. The responses of the unilateral
11 hearing-aid users did not differ significantly for any of the components P1, N1 or P2
12 between the aided and unaided ears, but a significant interaction between ear and
13 frequency was present for P2 amplitudes. P2 amplitudes were significantly smaller for
14 the 0.5- and 1-kHz stimuli and tended to be larger for the 2-kHz stimulus in the aided
15 ear suggesting an acclimatization effect. Larger P2 amplitudes were observed in the
16 unilaterally fitted group, which was interpreted as a correlate of more effortful auditory
17 processing in unilaterally fitted people.

18

19 **Keywords:** Acclimatization, AEP, auditory evoked potentials, bilateral, deprivation,
20 hearing aids; P2, unilateral

21

22 **Abbreviations:** ABR, auditory brainstem response; AEP, auditory evoked potential;
23 ANOVA, analysis of variance; DLF, discrimination limen for frequency; DLI, difference
24 limen for intensity; ENT, ear nose and throat; ISI, interstimulus interval; PTA, pure tone
25 average; S/N ratio, signal-to-noise ratio; SPIN, speech perception in noise; ULL,
26 uncomfortable loudness level.

27 **Introduction**

28 Hearing aids may be fitted to one ear only or to both ears. Bilateral provision has
29 become the standard for people with symmetrical hearing loss, because two hearing
30 aids are thought to be superior to one for most individuals. The possible benefits of
31 bilateral fitting comprise better speech understanding (Kobler et al., 2002; Moore et al.,
32 1992), in particular in noisy environments (Dreschler et al., 1994; Leeuw et al., 1991;
33 Nabelek et al., 1981), better sound quality (Balfour et al., 1992; Erdman et al., 1981),
34 better sound localization (Byrne et al., 1992; Dreschler et al., 1994; Kobler et al., 2001;
35 Punch et al., 1991; Stephens et al., 1991), and improved perception of distance and
36 movement (Noble et al., 2006). Principles of acoustics and hearing physiology also
37 support the use of bilateral fitting.

38 Furthermore, when a hearing aid is fitted in people with bilateral hearing loss to only
39 one ear, a large subset of people experience auditory deprivation in the unaided ear,
40 which is manifested as a significant reduction in speech recognition performance in the
41 unaided ear over time (Gelfand et al., 1987; Silman et al., 1984). This effect appears in
42 general after two to three years of deprivation (Arlinger et al., 1996). In the aided ear,
43 by contrast, an acclimatization effect may be observed, which was defined as an
44 improvement in auditory performance that cannot be attributed to training effects only
45 (Arlinger et al., 1996). During the Eriksholm workshop on auditory deprivation and
46 acclimatization, areas for future research for a better understanding of this
47 phenomenon were identified (Arlinger et al., 1996; Neuman, 1996). One of the
48 suggestions was to use electrophysiological and imaging techniques to understand the
49 anatomical and physiological changes underlying the mechanisms of deprivation and
50 acclimatization. Objective measures of the effects of bilateral versus unilateral fittings
51 on auditory processing may also be desirable, because clinical field studies have failed
52 to show a clear advantage of bilateral fitting (Noble et al., 2006).

53 Gatehouse (one of the workshop participants) and Robinson (1996) investigated
54 acclimatization to unilateral hearing-aid use in a single long-term user using simple
55 electrophysiological measures. The subject was a 69-year-old man who had been
56 aided in the right ear for 4 years with an average daily use of 8 hours. Auditory evoked
57 potentials (AEPs) were acquired for 500 and 2000 Hz sinusoids at three presentation
58 levels (65, 80, 95 dB SPL) for the aided and unaided ear separately. The composite
59 average of the N1-P2 amplitude was measured. For the lower frequency there was no
60 difference in N1-P2 amplitude between the ears at all levels, but for the 2000-Hz
61 stimulus, the aided ear had a larger amplitude for the 95 dB SPL presentation level.
62 The authors concluded that these results support a potential acclimatization effect
63 induced by the hearing aid and that future research could use more refined
64 electrophysiological measures to investigate changes induced by unilateral fitting.

65 Despite the suggestions of the Eriksholm workshop, studies using electrophysiological
66 measures to investigate the potential deprivational effects of unilateral hearing-aid use
67 are scarce. Munro et al. (2007a) investigated ear asymmetry in the auditory brainstem
68 response (ABR) of long-term unilateral hearing-aid users (minimum experience 2
69 years, self-reported daily use >5 hours) and a group of people with symmetric high-
70 frequency sensorineural hearing loss prior to hearing-aid fitting. Clicks were presented
71 unilaterally at 70, 80 and 90 dB HL. Wave V amplitudes were higher for the 70 and 80
72 dB levels in the aided ear compared to the unaided ear, which was interpreted as an
73 acclimatization effect at the brainstem level.

74 Hutchinson and McGill (1997) used P300, a discriminatory potential that is elicited in an
75 oddball paradigm by a rare stimulus presented randomly among a sequence of
76 frequent stimuli, to investigate auditory deprivation in ten unilaterally aided children
77 aged 9 to 18 years (mean 13.1 years). These children had bilateral congenital severe
78 to profound sensorineural hearing loss and had worn their aid for at least 8 years.

79 Stimuli (1000 Hz frequent, 250 Hz rare) were presented at a comfortable listening level
80 (varying between 80 and 118 dB nHL) for each subject. The P300 amplitude was
81 significantly greater in the aided ear compared to the unaided ear. Thus, the few
82 studies that used electrophysiological measures to investigate the effects of unilateral
83 hearing-aid use all reported increased amplitudes in the aided ear compared to the
84 unaided ear. Interestingly, this increase was observed across the whole range of
85 evoked potentials from ABRs to P300.

86 Evidence for acclimatization in unilateral hearing-aid users also comes from studies
87 investigating speech perception, intensity discrimination and loudness perception (for a
88 review, refer to Munro 2008). For example, Gatehouse (1989) used a speech-
89 perception-in-noise test, with a group of unilaterally fitted adults. He found better
90 performance at high presentation levels and poorer performance at low presentation
91 levels for the aided ear compared to the unaided ear. Intensity discrimination was used
92 by Robinson and Gatehouse (1995, 1996) to investigate acclimatization following
93 unilateral fitting. The difference limen for intensity (DLI) was measured at 0.25 and 3
94 kHz. In the aided ear, the DLI for the 3-kHz stimulus was better at high presentation
95 levels and poorer at low presentation levels. These results correspond to the findings of
96 Gatehouse (1989) regarding speech perception. Another measure used to investigate
97 acclimatization is the determination of uncomfortable loudness levels (ULL). Gatehouse
98 and Robinson (1996) and Munro et al. (2007b) found a greater tolerance of loudness in
99 the aided ear at higher frequencies (2 and 4 kHz). To summarize, all of these studies
100 present converging evidence that unilateral hearing-aid use improves perception in the
101 aided ear at high presentation levels, whereas performance at low presentation levels
102 tends to decrease. This effect was observed only for higher frequencies (≥ 2 kHz) and
103 for speech stimuli.

104 The current study was conducted for two purposes. First, we wanted to investigate the

105 effects of long-term unilateral hearing-aid use on late auditory evoked potentials. As a
106 first exploratory approach and using a study design similar to that of the case study by
107 Gatehouse and Robinson (1996), late AEPs comprising the P1-N1-P2 complex were
108 measured in a group of unilateral hearing-aid users. We hypothesized that unilateral
109 fitting would alter the responses. Specifically, we expected larger amplitudes and
110 shorter latencies of either all or some of the components P1, N1 and P2, when the
111 stimuli were presented to the aided ear compared to the responses from the unaided
112 ear.

113 A second purpose was to examine differences in amplitudes and latencies of P1, N1
114 and P2 across a group of unilateral hearing-aid users, age-matched bilateral hearing-
115 aid users and normal-hearing controls. These results are reported first. The nature of
116 this investigation was exploratory and no specific hypotheses were advanced.

117

118 **Materials and methods**

119 ***Participants***

120 Ten elderly bilateral hearing-aid users (mean age = 69.5 years, range 65-75 years;
121 eight men), 10 unilateral hearing-aid users (mean age = 77.1 years, range 73-86 years;
122 seven men) and a control group of 10 normal-hearing subjects (mean age = 70.1
123 years, range 66-73 years; 6 men) participated in the study. The unilateral users were
124 significantly older than the bilateral users ($t=4.58$; $p<0.001$) and the normal-hearing
125 subjects ($t=4.79$; $p<0.001$). Of the unilateral hearing-aid users, five wore their aid in the
126 right ear and five in the left ear. Given the small subsample size, loss of hemispheric
127 asymmetry, another potential consequence of unilateral amplification described in
128 people with unilateral hearing loss, was not addressed in our study. As asymmetry
129 seems to be affected differently by input from left and right ears (Hanss et al., 2009;
130 Hine and Debener, 2007; Thai-Van et al., 2009), the study did not have enough power

131 for this type of investigation. Only long-term hearing-aid owners with regular use,
132 defined as a total duration of at least 5 years and a daily self-reported use of ≥ 8 hours
133 per day, were included. The average duration of hearing-aid use was 6.3 years (SD 1.9
134 years, range 5-11 years) for the unilateral group and 12.4 years (SD 7.3 years, range
135 5-30 years) for the bilateral group. There was a significant difference of 6.1 years in
136 hearing-aid use duration between the two groups ($t=2.56$; $p<0.001$).

137 All hearing-aid users had digital aids with nonlinear signal processing features fitted by
138 professional hearing-aid dispensers according to the Swiss hearing-aid dispensing
139 system (Bertoli et al., 2009). No information about the real-ear insertion gain, i.e. the
140 difference between aided and unaided ear canal sound pressure level, was available.
141 However, the mean hearing threshold differences between 2 and 1 kHz were 18.3 dB
142 (SD 9.5 dB) and 16.3 dB (SD 10.9 dB), and between 2 and 0.5 kHz 22.0 dB (SD 12.2
143 dB) and 21.3 dB (SD 12.8 dB) for the right and left ear, respectively. It can therefore be
144 assumed that the hearing-aid gain was higher at 2 kHz compared to 0.5 and 1 kHz.

145 The hearing-impaired participants had a moderate high-frequency sensorineural
146 hearing loss with a pure-tone average (PTA) at 0.5, 1, 2, and 4 kHz between 40 and 60
147 dB HL. Hearing thresholds did not differ significantly between unilateral and bilateral
148 hearing-aid users. Hearing loss had to be symmetrical with a PTA difference between
149 left and right ears not exceeding 10 dB. No significant differences between the hearing
150 thresholds of right and left ears were found for any of the frequencies in both hearing-
151 impaired groups (p -values between 0.09 and 0.84). Otoscopy and acoustic immittance
152 testing were used to control for conductive hearing loss. The normal-hearing group had
153 a PTA of 20 dB HL or better in both ears. The mean pure-tone audiograms of the three
154 groups for the right and left ears are depicted in Figure 1. In addition, all subjects
155 passed a screening test for dementia using a German version of the
156 neuropsychological assessment battery of the Consortium to Establish a Registry for

157 Alzheimer's Disease (CERAD-NAB) with normative values adjusted for gender, age,
158 and education (Thalman et al., 2000; Welsh et al., 1994).

159 Hearing-aid users were recruited among participants of a prior study (Bertoli et al.,
160 2010; Bertoli et al., 2009), from local hearing-aid dispensers and from the ENT-
161 department of the University Hospital Basel. The normal-hearing subjects had either
162 participated in prior studies or were recruited from a local longitudinal study on healthy
163 aging. The study was approved by the local Ethics Committee of Basel and Baselland
164 (EKBB) and all participants gave written informed consent prior to testing.

165

166 ***Speech audiometry***

167 Two measures of speech perception were used to investigate whether perceptual
168 evidence for acclimatization had occurred in the aided ear compared to the unaided ear
169 in the unilateral hearing-aid users. The 50% correct speech recognition for
170 monosyllabic words was determined using the Freiburger Einsilbertest (Hahlbrock,
171 1953). A modified German version of the speech-perception-in-noise (SPIN) test was
172 administered using the sentences with low predictability to determine the signal-to-
173 noise (S/N) ratio for which 50% of the final words of sentences presented in a constant
174 background noise are correctly identified (Kalikow and Stevens, 1977; Tschopp and
175 Züst, 1994). For the normal-hearing control group, the noise level was set at 60 dB
176 SPL. For the hearing-impaired participants, the noise level was calculated by adding 30
177 dB to the 50% speech recognition score.

178

179 ***Stimuli and electrophysiological procedure***

180 The stimuli were 0.5, 1 and 2 kHz pure-tones with a duration of 100 ms and a 10-ms
181 rise/fall time. They were presented at 55, 70 and 85 dB SPL via ER3 insert earphones

182 either to the right or left ear, resulting in a total of 18 conditions (3 frequencies x
183 3 levels x 2 ears). Stimuli were delivered with an interstimulus interval (offset-to-onset)
184 of 1s in two separate blocks of 900 stimuli each. Each block contained 50 presentations
185 of each stimulus type and the order in which the stimuli were presented varied between
186 the two blocks. Thus, each stimulus type was presented 100 times. The duration of one
187 test block was about 20 min.

188 Recordings were conducted in a sound-treated and electrically shielded room.
189 Participants were instructed to ignore the sounds and to concentrate on reading a text
190 of their own choice.

191

192 ***EEG recording and averaging***

193 The EEG was recorded using a Neuroscan Quicktrace system and disposable surface
194 silver electrodes at Fz, Cz, Pz, left and right mastoids (LM, RM) according to the
195 International 10/20 system, and at two lateral sites halfway between Fz - LM and Fz -
196 RM, respectively (L1, R1). An electrode placed at the tip of the nose served as the
197 reference and a forehead electrode as ground. Vertical eye movements were
198 monitored with two electrodes attached above and below the left eye. Impedance was
199 kept below 5 k Ω and controlled between the two test blocks.

200 The EEG (band pass 0.05 – 100 Hz) was recorded continuously at a sampling rate of
201 500 Hz and stored for off-line averaging. An ocular artifact reduction algorithm was
202 used to reduce contamination by eye movements. Epochs containing 100-ms pre-
203 stimulus and 500-ms post-stimulus time were obtained, baseline-corrected with respect
204 to the pre-stimulus interval, and averaged by stimulus type. Epochs containing artifacts
205 exceeding ± 100 μ V were rejected from averaging. The AEP waves were band-pass
206 filtered at 0.1 – 20 Hz (24 dB/octave slope).

207

208 ***Electrophysiological data analysis***

209 For each subject, events corresponding to each condition were averaged. Grand mean
210 average waveforms were calculated for each subject group and stimulus type. The P1,
211 N1 and P2 peak amplitudes and latencies were measured in the waveforms at Cz,
212 where the largest potentials were seen, and at Pz, because there were clear responses
213 in the unilateral group that were less prominent or absent in the other two groups. The
214 composite N1-P2 amplitude was also calculated. The latency windows for the peak
215 measurements were determined based on the grand average waveforms (P1: 20-90
216 ms, N1: 40-170 ms, P2: 120-340 ms). In addition, to account for the sustained and
217 double-peaked P2, two mean amplitude voltages were measured for the 130 - 240 and
218 the 240 – 350 ms latency ranges (mean P2_{early} and mean P2_{late}, respectively). To
219 correct for multiple comparisons (two electrode sites), alpha level was adjusted to
220 <0.025.

221 Data were analyzed using SPSS software (version 19). The P1, N1 and P2 amplitudes
222 and latencies were analyzed using separate repeated-measures ANOVAs for electrode
223 sites Cz and Pz with subject group as the between-subject factor (unilateral, bilateral,
224 normal) and ear (left, right), frequency (0.5, 1, 2 kHz), and level (55, 70, 85 dB SPL) as
225 within-subject factors. Huynh-Feldt corrections were used where an assumption of
226 sphericity was not appropriate. When significant main effects were found for subject
227 group or interactions, Bonferroni's post-hoc measures were performed (alpha level
228 <0.05). Significant main effects for frequency and level were not further investigated
229 with post-hoc analyses, since the effects of these parameters on AEPs have been
230 studied extensively in the past (for a review, see Crowley and Colrain, 2004; Hyde,
231 1997) and were not of specific interest for the purpose of the current study.

232 To investigate the effect of hearing-aid use on AEPs, the ears of the unilateral group

233 were classified as aided and unaided. For bilateral users, ears were classified as left
234 and right. Difference values for the two ears were calculated for all parameters (P1, N1,
235 P2 amplitudes and latencies). For the unilateral group, results for the unaided ear were
236 subtracted from those of the aided ear, for the bilateral group right ear results were
237 subtracted from those of the left ear. Repeated-measures ANOVAs were then
238 calculated for the difference values with factors subject group (unilateral, bilateral),
239 frequency and level. The group with normal hearing was not included in this analysis.
240 To investigate differences between aided and unaided ears in the unilateral group
241 further, repeated-measures ANOVAs were performed with factors ear, frequency and
242 level for N1 and P2 amplitudes and for the composite N1-P2 amplitude to enable a
243 direct comparison of our results with those of the case study by Gatehouse and
244 Robinson (1996). This analysis was performed for the unilateral group only.

245

246 **Results**

247 ***Speech audiometry***

248 The results of the 50% speech discrimination and SPIN tests are listed in Table 1. For
249 the normal-hearing and bilateral groups, results are reported for left and right ears, for
250 the unilateral group for aided and unaided ears. The hearing-aid users' performance
251 was significantly poorer on both tests compared to the normal-hearing people (p -values
252 <0.001). Speech performance of the aided and unaided ears of the unilaterally fitted
253 group was compared to each ear (right and left) of the bilaterally fitted group. None of
254 the comparisons for the 50% speech discrimination and S/N ratio reached significance
255 (p -values between 0.09 and 0.95). In the unilateral group, the 50% speech
256 discrimination scores between aided and unaided ears did not differ significantly
257 ($p=0.105$), although there was a trend towards better scores for the unaided ear. For
258 the SPIN test, the S/N ratio was significantly better in the aided ear compared to the

259 unaided ear ($p=0.012$). In the bilateral and normal-hearing groups, no significant
260 differences were noted between right and left ears for any of the speech tests.

261

262 ***AEP results***

263 ***Comparison by subject group***

264 ***Visual inspection of the waveforms***

265 Figure 2 displays the responses to the 0.5 kHz stimulus presented at 85 dB SPL to the
266 left ear at all eight electrode sites. Figure 3 depicts the grand mean average waveforms
267 of the three subject groups for all 18 conditions at electrode sites Cz and Pz. For the
268 normal-hearing group, the typical P1-N1-P2 complex was present for all frequencies
269 and intensity levels at electrode site Cz. The hearing-aid users had clear responses for
270 all stimulus types except the 2 kHz-tone presented at 55 dB SPL. For most hearing-
271 impaired participants, this stimulus was below their hearing thresholds. At Pz, the group
272 with unilateral hearing-aid provision had clearly visible responses for the 0.5 and 1 kHz
273 stimuli, in particular at 70 and 85 dB SPL presentation level, whereas the responses
274 were considerably reduced or absent in the bilateral and normal groups. At the lowest
275 presentation level, the normal-hearing participants had larger N1 amplitudes compared
276 to the hearing-impaired participants, whereas at the higher levels only minor
277 differences for the P1 and N1 components could be noted. A pronounced and
278 sustained P2 was found in the responses of the unilateral group compared to the
279 bilateral and normal groups, who had generally smaller P2s. This effect was more
280 pronounced at Pz. In some of the waveforms, P2 was double-peaked.

281

282 ***Amplitudes***

283 Table 2 presents the results of the repeated-measures ANOVAs performed for P1, N1,

284 and P2 peak amplitudes and latencies and P2 mean voltages. There was a significant
285 main effect of subject group at Pz on P2 peak amplitude and on the early portion of the
286 mean P2 voltage, but not on N1 amplitude. Post-hoc analyses indicated that the
287 unilateral group had significantly larger amplitudes than the bilateral group (P2 peak:
288 $p=0.009$; mean P2_{early}: $p=0.007$). The means of the individual P2 amplitudes are plotted
289 in Figure 4 for electrode sites Cz and Pz.

290 There was a significant main effect of level on the amplitudes of all components (P1,
291 N1, P2), indicating larger amplitudes for the higher stimulus levels. Frequency affected
292 N1 and P2 amplitudes and the mean P2_{early} significantly, indicating smaller amplitudes
293 for higher frequencies. No significant interactions of subject group with ear, frequency
294 and level were observed for any of the AEP components.

295

296 ***Latencies***

297 There was a significant main effect of subject group on P1 latency at Cz. Both groups
298 of hearing-aid users had longer latencies compared to the normal group (unilateral
299 47 ms, bilateral 49 ms, normal 42 ms). Post-hoc tests revealed that the differences
300 were significant only for the bilateral group ($p=0.014$). There was also a trend towards a
301 significant group effect on N1 latency at Cz ($F(2,27)=3.59$; $p=0.041$) with prolonged
302 latencies for the two hearing-aid user groups (unilateral 105 ms, bilateral 108 ms,
303 normal 100 ms). P2 latency was not affected significantly by subject group.

304 Frequency affected P1 and N1 latencies at Cz and Pz, whereas level affected N1
305 latency at Cz and P2 latency at Pz significantly. Again, no significant interactions of
306 subject group with ear, frequency and level were observed for any of the AEP
307 components.

308

309 ***Effects of unilateral vs. bilateral fitting***

310 Figure 5 depicts the responses for all frequencies and levels for the two hearing-aid
311 user groups at electrode site Cz. For the bilateral group, results are plotted for the right
312 and left ear, whereas for the unilateral group results are plotted for the aided and
313 unaided ear. In Figure 6, the means of the individual peak amplitudes and latencies are
314 displayed for P1, N1 and P2.

315 Repeated measures ANOVAs showed a significant main effect of frequency on P2
316 amplitude differences between the two ears at Cz. For the frequencies 0.5 and 1 kHz,
317 the average amplitude difference values were negative (-0.25 and -0.40 μV), whereas
318 for 2 kHz the difference value was +0.25 μV . Post-hoc tests indicated a significant
319 difference between the frequencies 1 and 2 kHz ($p=0.001$). There was no significant
320 main effect of group and level nor was there an interaction between the factors for any
321 of the parameters investigated indicating that unilateral hearing-aid use did not affect
322 the responses differently compared to bilateral use. Table 3 summarizes the results of
323 the repeated measures ANOVAs.

324 Despite the lack of a significant main effect of subject group, visual inspection of the
325 plots for the mean P1, N1 and P2 amplitudes in Figure 6 revealed diverging trends for
326 P2 amplitudes of aided and unaided ears in the unilaterally fitted group not observed in
327 the bilaterally fitted group. Divergence increased with increasing presentation level in a
328 frequency-specific manner. For the 0.5-kHz and 1-kHz stimuli, P2 amplitudes appeared
329 larger in the unaided ear, and for the 2-kHz stimulus, P2 amplitudes appeared larger in
330 the aided ear. To investigate these trends further, additional ANOVAs were performed
331 for the unilateral group alone with factors ear, frequency and level. For comparison
332 purposes with the study of Gatehouse and Robinson (1996), the ANOVAs were also
333 performed for the composite N1-P2 in addition to N1 and P2 amplitudes. Results are
334 given in Table 4. There was a significant main effect of level (at Cz and Pz) and of

335 frequency (Cz only), but not of ear, and there was a significant interaction between ear
336 and frequency for P2 peak amplitude (at Cz) and for the composite N1-P2 amplitude
337 (at Pz). Post-hoc tests revealed that P2 amplitudes were significantly larger in the
338 unaided compared to the aided ears at the frequencies 0.5 kHz ($p=0.044$) and 1 kHz
339 ($p=0.026$), but P2 tended to be smaller in the unaided ear at 2 kHz ($p=0.170$). The
340 composite N1-P2 amplitudes were significantly larger in the unaided ear for the 1-kHz
341 stimulus ($p=0.020$).

342 The trend for a larger P2 amplitude in the aided ears at 2 kHz is consistent with the
343 larger N1-P2 amplitude in the aided ear at the same frequency reported in the case
344 study of Gatehouse and Robinson (1996). Unlike Gatehouse and Robinson (1996), we
345 found also an acclimatization effect at the lower frequencies of 0.5 and 1 kHz with
346 significantly larger P2 amplitudes in the unaided ears. Our results also show that the
347 acclimatization effect is related to changes in P2 amplitude and not in N1 amplitude.

348

349 **Discussion**

350 This study examined the hypothesis of an acclimatization effect on electrophysiological
351 measures in the aided ear of experienced unilateral compared to bilateral hearing-aid
352 users and a normal-hearing control group. In the unilateral group, no significant
353 differences in P1, N1 and P2 amplitudes and latencies were found between the
354 responses obtained separately from the aided and unaided ears. There was, however,
355 a significant interaction between ear and frequency with smaller P2 amplitudes for the
356 0.5- and 1-kHz stimuli and a trend towards larger P2 amplitudes for the 2-kHz stimulus
357 in the aided ear. Using speech audiometry, significantly lower S/N ratios for the aided
358 ear compared to the unaided ear were demonstrated. Thus, the current study provides
359 some electrophysiological and perceptual evidence for an acclimatization effect in the
360 aided ears of unilateral hearing-aid users.

361 A comparison of the AEPs between unilateral and bilateral hearing-aid users and a
362 group of elderly normal-hearing people revealed no significant findings except for larger
363 P2 amplitudes in the unilateral group compared to the bilateral group, and longer P1
364 and N1 latencies in both hearing-aid groups compared to the normal-hearing group, but
365 this increase was significant only for the bilaterally fitted group.

366

367 ***Acclimatization effects on late AEPs with unilateral hearing-aid use***

368 An acclimatization effect of unilateral amplification has been reported using
369 electrophysiological measures with widely different latencies such as short latency ABR
370 and long latency P300 (Hutchinson and McGill, 1997; Munro et al., 2007a). Munro et
371 al. (2007a) reported larger wave V amplitudes in the aided ear compared to the
372 unaided ear. Investigating the effect of bilateral hearing-aid fitting on ABR, Philibert et
373 al. reported significantly shortened wave V latencies only in the right ear, but not in the
374 left ear after 3 and 6 months of regular bilaterally used hearing aids (Philibert et al.,
375 2005). These ABR results suggest that acclimatization effects may occur at the initial
376 more peripheral stages of central auditory processing. However, in both ABR studies
377 only a small number of participants were tested (eight and five, respectively) and
378 significant results were inconsistent and limited to either amplitude or latency of wave V
379 or to one side only.

380 Hutchinson and McGill (1997) reported significantly greater P300 amplitudes in the
381 aided ears of unilaterally aided children. Although these children had worn their hearing
382 aids for at least 8 years, methodological differences preclude a direct comparison. First,
383 the children had congenital severe to profound hearing loss. Second, at the age of 9 to
384 17 years long latency AEPs are still subject to maturational changes. Third, the P300 is
385 a discriminative potential that requires attention to the stimuli.

386 Our study can be compared more readily to the results of Gatehouse and Robinson
387 (1996). These authors reported a potential acclimatization effect in a single subject for
388 the highest presentation level of the 2-kHz stimulus with larger N1-P2 amplitudes for
389 the aided ear, but not for the 0.5-kHz stimulus. As Gatehouse and Robinson had
390 measured the composite N1-P2 amplitude, it is unknown whether this difference was
391 related to changes in N1, P2, or both. In the current study, we found a significant
392 interaction between ear and frequency in the unilateral group for P2 and the composite
393 N1-P2 amplitude, but not for N1. Inspection of Figures 5 and 6 revealed diverging
394 trends for P2 in the aided and unaided ears with increasing levels, depending on the
395 test frequency, whereas no such trends can be noted for N1. The results of the current
396 study are consistent with the results of Gatehouse and Robinson (1996) for the 2-kHz
397 stimulus and suggest that the changes are specifically due to an increase in P2
398 amplitude. In addition, unlike Gatehouse and Robinson (1996), who did not find
399 differences between aided and unaided ears for the 0.5-kHz stimulus, our results
400 extend the finding of an acclimatization effect to the lower frequencies of 0.5 and 1 kHz
401 with larger P2 amplitudes at higher levels in the unaided ear. This means that
402 acclimatization occurs at higher presentation levels at the frequencies to which the ear
403 is most exposed: the unaided ear to the lower frequencies, and the aided ear to the
404 higher frequencies. The fact that similar changes are not present in the bilaterally fitted
405 group suggests that the changes are related to the asymmetry in the listening
406 conditions across ears in the unilateral group.

407 The frequency specificity of the P2 enhancement with larger amplitudes in the unaided
408 ear at 0.5 and 1 kHz and larger amplitudes in the aided ear at 2 kHz could also be
409 interpreted as a mild form of representational plasticity. Responsiveness to the edge
410 frequencies of dead regions has been reported for neurons adjacent to such regions. It
411 is possible that with presbycusis, neural resources previously tuned to high-frequency

412 input become responsive to lower frequency input. Some evidence of such changes
413 following gently sloping hearing losses have been reported in animal models (e.g.,
414 Frisina and Rajan, 2005). Moreover, several studies in humans have demonstrated that
415 patients with steeply sloping sensorineural hearing loss exhibit an improvement in
416 frequency discrimination performance at or around the cut-off frequency (McDermott et
417 al., 1998; Thai-Van et al., 2002, Thai-Van et al., 2007). Following the introduction of a
418 hearing aid, it might be that the neural resources shift back to respond to higher
419 frequency input once more. This would explain the larger P2 amplitudes for low
420 frequencies in the unaided ear and the larger P2 amplitudes for high frequencies in the
421 aided ear. Gabriel et al. (2006) have demonstrated the existence of such a secondary
422 plasticity induced by auditory rehabilitation. The discrimination limen for frequency
423 (DLF) was investigated at the frequency with the best DLF before and at 1, 3 and 6
424 months following hearing-aid fitting. The DLF of the best frequency decreased
425 significantly, while remaining stable at other frequencies. This change was interpreted
426 as a central reorganization induced by amplification and reversing the initial hearing-
427 loss induced changes in the cortical maps.

428

429 ***Speech audiometry in unilateral and bilateral hearing-aid users***

430 When speech performance was compared between the ears of unilateral and bilateral
431 long-term hearing-aid users, no significant differences were found. When the ears were
432 compared within the unilateral group alone, S/N-ratios were significantly better for the
433 aided compared to the unaided ear, but not speech discrimination scores. Thus, some
434 audiometric evidence of acclimatization in unilateral hearing-aid users could be noted.
435 Whether the worse S/N ratio in the unaided ear represents simply the lack of
436 acclimatization or an additional deprivation effect (= deterioration compared to
437 performance before wearing a hearing aid), cannot be deduced from our study due to

438 its cross-sectional design. The SPIN-test was presented to the hearing-impaired
439 subjects at constant noise levels above 80 dB SPL (see Table 1). Our results are in line
440 with those of Munro and Lutman (2003), who reported an acclimatization effect after 12
441 weeks of hearing-aid use, when using speech in noise at the highest presentation level
442 of 69 dB SPL, but only minimal for 55 and 62 dB SPL. Gatehouse (1989) also reported
443 that the aided ears performed better only at high presentation levels (>75 dB SPL),
444 while at lower presentation levels the unaided ear was advantaged. The lack of a
445 significant difference between aided and unaided ears for the speech discrimination
446 test might therefore be related to the lower presentation levels between 53 and 57 dB
447 SPL (see Table 1). Alternatively, the SPIN-test representing a more complex listening
448 situation might be better suited for revealing acclimatization and deprivation effects
449 than simple speech discrimination tasks.

450

451 ***P1 and N1 latency increase in hearing-aid users***

452 The hypothesis that deprivation should cause changes in AEPs is partially based on
453 the finding that sensorineural hearing loss is associated with a prolongation of latencies
454 of the P1 and N1 components, occasionally also of P2 (Bertoli et al., 2005; Korczak et
455 al., 2005; Oates et al., 2002; Polen, 1984; Tremblay et al., 2003). Oates et al. (2002)
456 tested adults with hearing losses ranging from mild to severe and found prolonged ERP
457 latencies with even mild hearing loss, whereas amplitudes were affected only in
458 participants whose average hearing loss exceeded 60 dB HL. They suggested that any
459 sensorineural hearing loss results in an overall slowing of the timing of the cognitive
460 processes.

461 In accordance with these findings, the two groups of participants with symmetrical
462 hearing loss and hearing-aid use showed a prolongation of the latencies of P1 and N1
463 compared to the normal-hearing group, but not of P2. The finding that P2 latency was

464 not significantly prolonged despite the latency increases of the preceding components
465 P1 and N1 could be related to the sustained P2 latency range with a large variability of
466 P2 peak amplitude measures.

467

468 ***P2 enhancement in unilateral hearing-aid users***

469 A significantly larger P2 amplitude at Pz was found in the unilateral group in our study
470 compared to the bilateral group. Unlike the N1 component of the late AEPs, the P2
471 component has not received much attention in the past, because it was considered to
472 reflect the same neural mechanisms as the preceding N1 (Crowley and Colrain, 2004).
473 As a consequence, data analysis was frequently limited to N1 or the composite N1-P2
474 amplitude and little is known about the functional significance of P2. Interpretations of
475 P2 findings in the literature are frequently of a speculative nature. In a review, Crowley
476 and Colrain (2004) documented that P2 can be dissociated from N1 experimentally,
477 developmentally and topographically. An enhanced P2 has been reported from other
478 areas of research, such as sleep (Crowley et al., 2002), dyslexic children (Ceponiene
479 et al., 2009), and from auditory discrimination training (Alain and Snyder, 2008; Atienza
480 et al., 2002; Bosnyak et al., 2004; Reinke et al., 2003; Ross and Tremblay, 2009; Tong
481 et al., 2009; Tremblay et al., 2001; Tremblay and Kraus, 2002; Tremblay et al., 2010)
482 as well as from mere passive exposure to repeated presentations of stimuli (Ross and
483 Tremblay, 2009; Sheehan et al., 2005; Tremblay et al., 2010). Larger P2 amplitudes
484 have also been associated with aging (Amenedo and Diaz, 1999; Ceponiene et al.,
485 2008), but the literature is inconsistent, reporting also diminished or unchanged P2
486 amplitudes (for a review see Ceponiene et al., 2008).

487 To understand the functional significance of our finding in a group of unilateral hearing-
488 aid users, it might be helpful to review the interpretations from different areas of
489 research reporting enhanced P2 amplitudes and identify processes that could be

490 similar to the listening experience of the hearing-aid users, such as auditory training
491 programs using discrimination or identification tasks to train their participants (Alain and
492 Snyder, 2008; Ross and Tremblay, 2009; Tremblay and Kraus, 2002; Tremblay et al.,
493 2009). All these studies reported an experience-related enhancement of P2 amplitudes
494 that has been interpreted as reflecting enhanced arousal or awareness of trained
495 stimuli. As P2 is thought to reflect the auditory-driven output of the mesencephalic
496 reticular activating system (Crowley and Colrain, 2004), perhaps the training activates
497 a preattentive alerting mechanism that contributes to improved perception.

498 The experience of a hearing-aid user that is exposed to new auditory stimuli made
499 available through the hearing aid resembles the experience of auditory discrimination
500 training, where new acoustic features are learned. In our study, only the unilaterally
501 fitted hearing-aid users had significantly larger P2 amplitudes, but not the bilaterally
502 fitted group. This finding suggests differences in the hearing experience of the two
503 hearing-aid. If P2 reflects a preattentive alerting mechanism, then its enhancement
504 could be related to the current hearing experience indicating that the unilateral group is
505 more alerted – even under passive and non-demanding listening conditions as in our
506 study – and directs more attention or processing resources to listening than normal-
507 hearing people and bilaterally fitted hearing-impaired people.

508 Admittedly, this interpretation is speculative due to the cross-sectional design of our
509 study without data on the dynamics of changes following hearing-aid fitting.

510 Longitudinal studies are needed that document the time course before hearing-aid
511 provision and during a follow-up period of months or even years to elucidate further the
512 plastic changes induced by amplification in the central auditory system.

513 A further characteristic of the P2 in our study was its scalp distribution with a significant
514 P2 enhancement in the unilateral group at electrode site Pz only (Fig. 4), whereas the
515 overall topographic distribution was similar for the three subject groups with maximum

516 values at Cz. In contrast, Tremblay and Kraus (2002) reported training-induced
517 increases in P2 amplitude that were significant over both hemispheres across all
518 midline and hemispheric recording sites. The authors questioned whether this
519 widespread distribution of change of P2 suggests global rather than specific acoustic
520 processes. The meaning of the parietal focus of P2 enhancement in the unilateral
521 group in our study is unknown. Perhaps it also points to more general and modality-
522 independent processes related to alertness and arousal.

523 Finally, an additional characteristic of the P2 in our study was the sustained double-
524 peaked nature of P2 that was found in all three subject groups and was clearly visible
525 in some traces (Figure 3). Our attempt to account for the double-peaked P2 by
526 quantifying two subsequent portions of P2 over the latency range of the sustained
527 positivity yielded a significant difference between the unilateral and bilateral groups for
528 the early portion only, but not for the late. The latency windows for the early and late
529 mean P2 amplitudes in our study were 130-240 ms and 240-350 ms, respectively. A
530 similar double-peaked P2 has been described by Ceponiene et al. (2008) in a study on
531 the effects of aging on auditory and visual processing. They found an age-related
532 enhancement for both modalities in the later P2 range with a peak at about 250 ms.
533 Regarding the auditory P2, they hypothesized that the late portion of P2 in the older
534 group was caused by an overlap with a positivity not identical to the auditory P2 but
535 similar to the positivity seen in the visual data. Ceponiene et al. (2008) did not provide
536 an interpretation for the functional significance of the hypothetical late auditory P2 and
537 the late visual P2 in the older group. However, the similarity of the auditory and visual
538 P2 findings suggests a modality-independent underlying process.

539 In our study, a significant difference was found only for the early portion of P2 and
540 between the two hearing-aid user groups. If the early P2 largely represents the "proper"
541 auditory P2, then the difference between unilateral and bilateral hearing-aid users

542 could be related to more specific auditory processing differences and not to global
543 modality-independent processes represented by the late portion of P2.

544 Even though the functional significance of the potentially two or more processes in the
545 130-350 ms latency window remains unknown, this latency range appears to be
546 sensitive not only to the effects of aging (Ceponiene et al., 2008), but also of sensory
547 acclimatization and deprivation. It may deserve more attention in future research.

548

549 ***Confounding factors***

550 Several potentially confounding factors must be taken into account in the interpretation
551 of our data. The unilateral group was older in age than the two other subject groups
552 (77.1 vs. 69.6 and 70.1 years), and their overall duration of hearing-aid use was shorter
553 than in the bilateral group (6.2 vs. 12.4 years). This imbalance reflects the prescription
554 practice in Switzerland with different reimbursement criteria for the retired and working
555 population. People who are employed are reimbursed for bilateral fittings, whereas
556 people who are retired are reimbursed for one aid only. It cannot be excluded that the
557 larger P2 of the unilateral group may be attributed partially to the older age rather than
558 being a specific consequence of the unilateral fitting, or to the shorter overall duration
559 of hearing-aid use. However, an aging effect is unlikely to account for the larger P2 in
560 the unilateral group compared with the bilateral group since this would have also
561 predicted a larger P2 in the unilateral group compared to the normal-hearing group,
562 which was not observed.

563

564 **Conclusions**

565 The current study used late AEPs to investigate acclimatization effects following long-
566 term unilateral hearing-aid use. A simple acclimatization effect with increased and

567 shorter responses for the aided ear compared to the unaided ear was not observed,
568 but for P2 there was a significant interaction between ear and frequency indicating
569 larger P2 amplitudes with the 2-kHz stimulus in the aided ear and larger P2 amplitudes
570 with the 0.5- and 1-kHz stimuli in the unaided ear suggesting the presence of
571 acclimatization. These results replicate and support the findings of Gatehouse and
572 Robinson (1996) from a single subject.

573 P2 was also the only component of the P1-N1-P2 complex that appeared sensitive to
574 capturing differences in central auditory processing between unilaterally and bilaterally
575 fitted people. The double-peaked nature and sustained latency range of P2 suggests
576 that two or more and possibly overlapping processes are contributing to this
577 component representing both modality-specific auditory and more global modality-
578 independent processes. The enhanced P2 amplitude in unilateral hearing-aid users in
579 our study was interpreted as a potential correlate of more effortful auditory processing
580 associated with the unilateral fitting compared to bilateral fitting. This interpretation is
581 somewhat speculative and future research should explore further the functional
582 significance of P2 in people with hearing impairment and try to untangle the presumed
583 various and overlapping processes in the latency range of the sustained P2
584 component.

585

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754 **Figure captions**

755

756 **Figure 1:** Mean hearing thresholds (± 1 standard deviation) of right and left ears for the
757 bilaterally and unilaterally fitted hearing-aid users and for the normal-hearing controls.

758 **Figure 2:** Grand average waveforms of the three subject groups (unilateral, bilateral,
759 normal) for the 0.5 kHz stimulus presented at 85 dB SPL to the left ear at all eight
760 electrode sites.

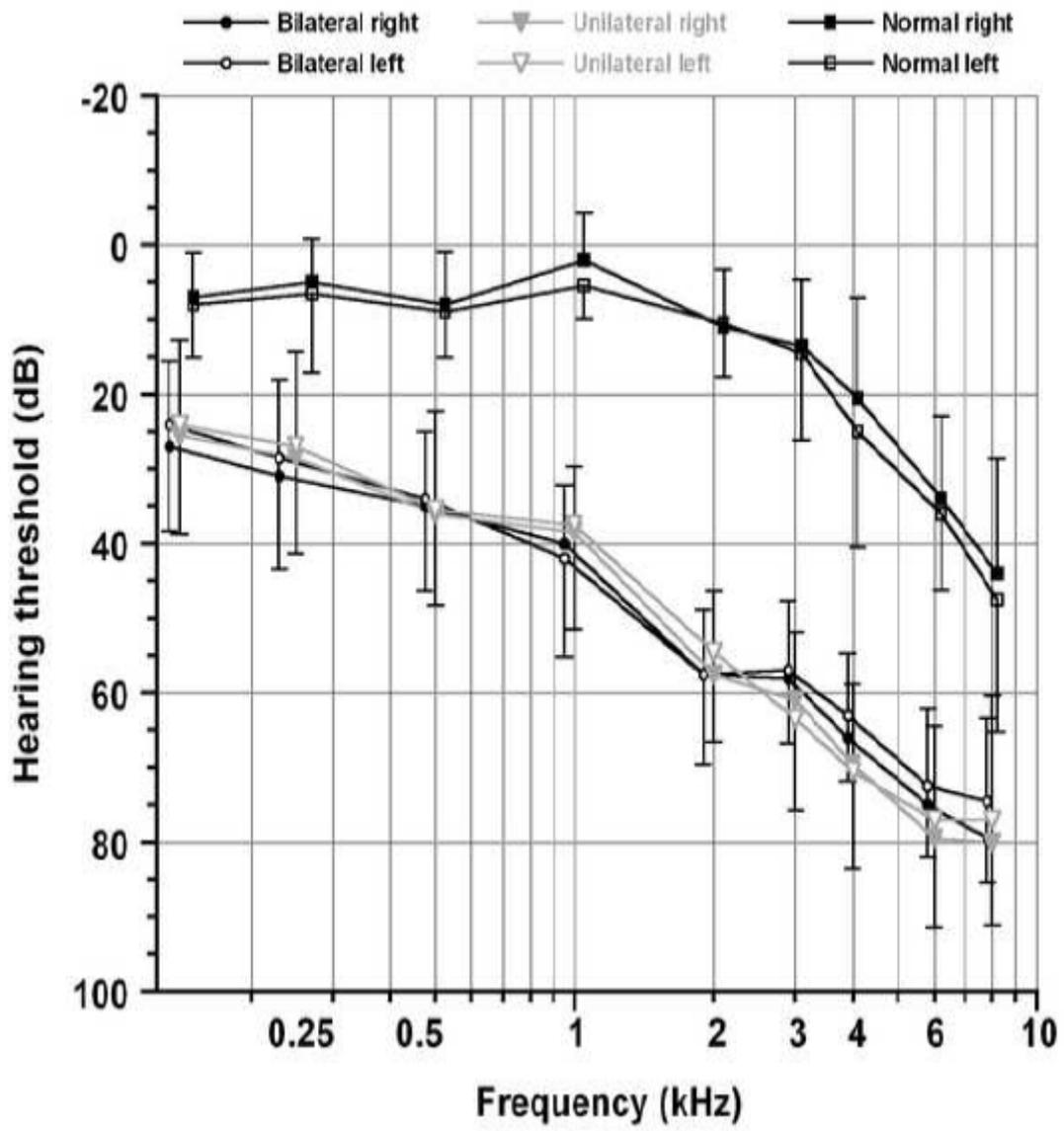
761 **Figure 3:** Grand average waveforms of the three subject groups (unilateral, bilateral,
762 normal) for all 18 conditions recorded at electrode sites Cz and Pz.

763 **Figure 4:** Mean P2 peak amplitudes (± 1 standard error) of the three subject groups
764 (unilateral, bilateral, normal) for the 18 conditions recorded at electrode sites Cz and
765 Pz.

766 **Figure 5:** Comparison of the AEPs from the aided versus unaided ear (for the unilateral
767 group) and from the left versus right ear (for the bilateral group) for the three
768 frequencies (0.5, 1, 2 kHz) and presentation levels (55, 70, 85 dB SPL) at electrode
769 site Cz.

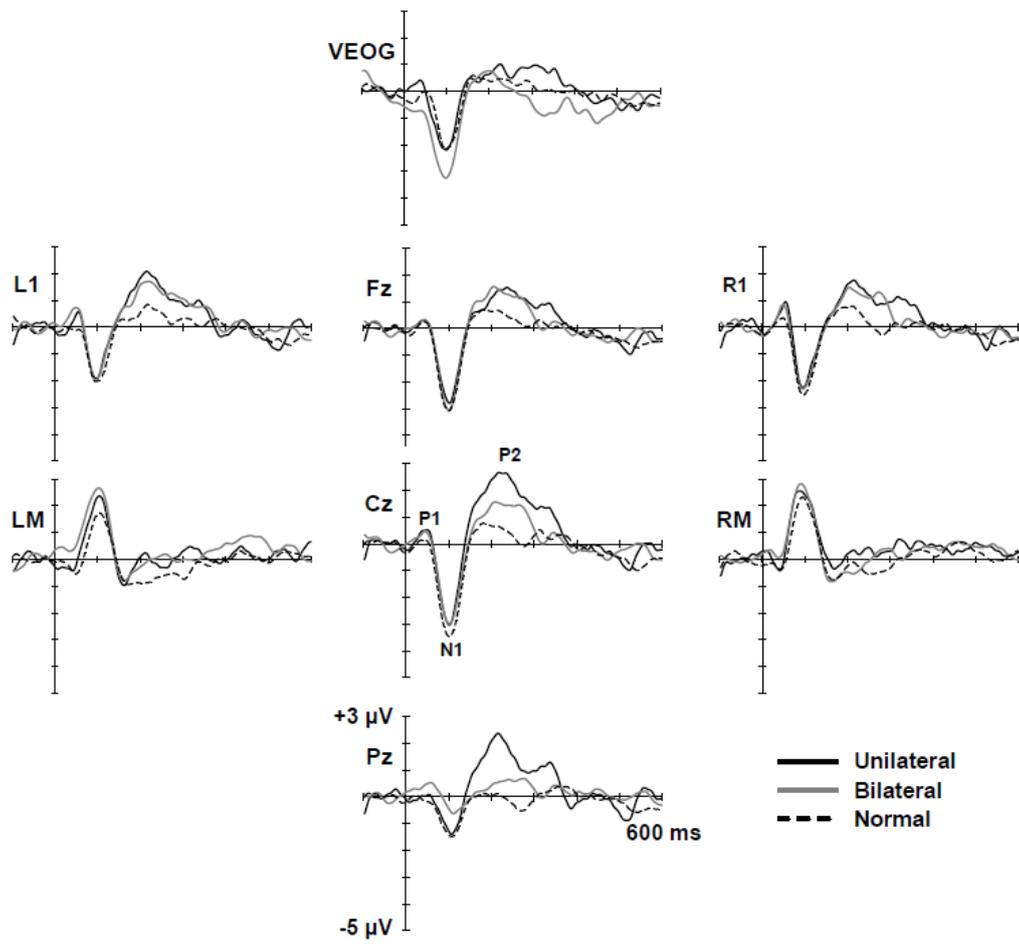
770 **Figure 6:** Mean P1, N1 and P2 peak amplitudes (left panel) and latencies (right panel)
771 at electrode site Cz plotted as a function of aided versus unaided ear (for the unilateral
772 group) and left versus right ear (for the bilateral group).

773 Figure 1:



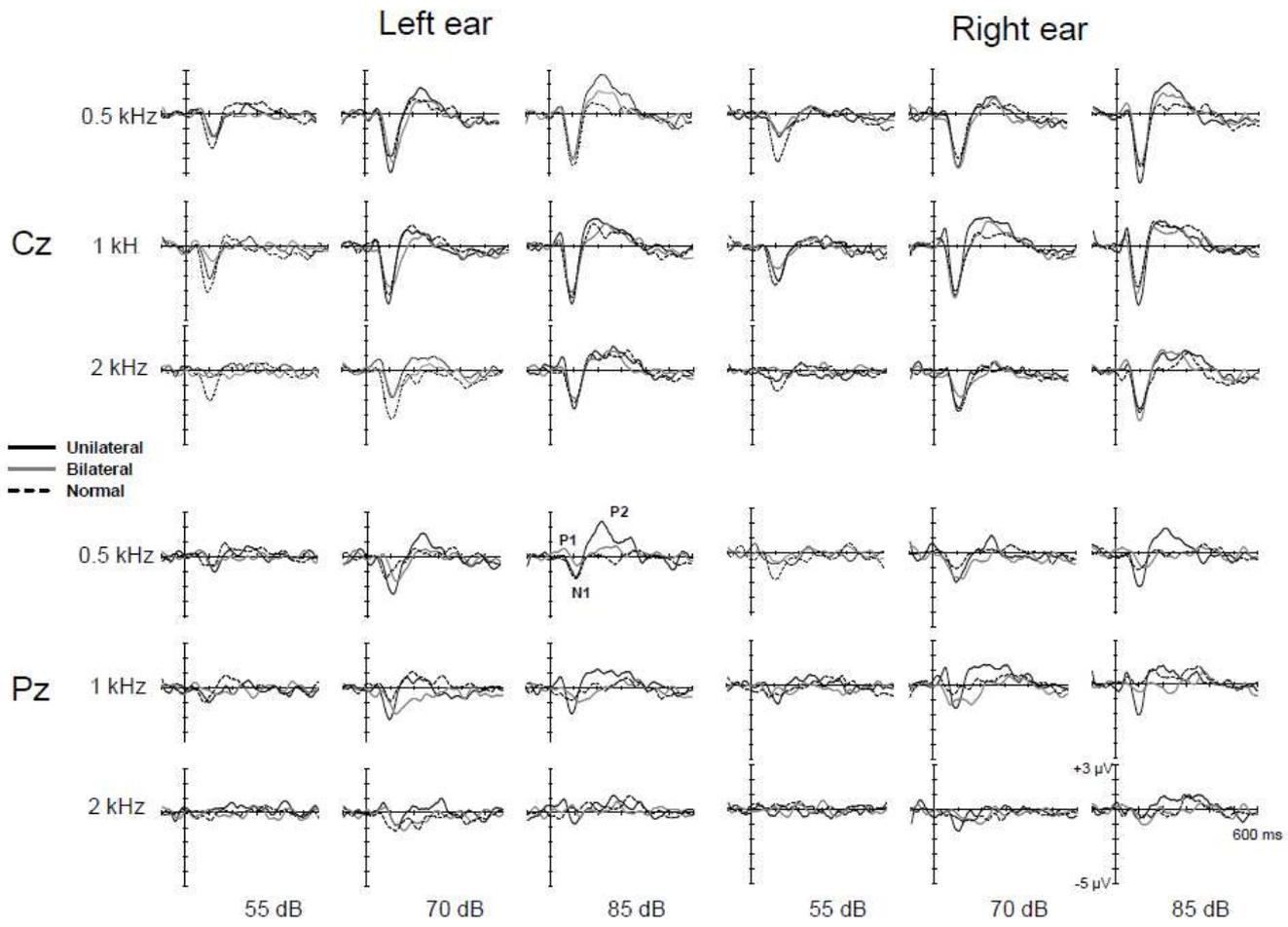
774

775 Figure 2



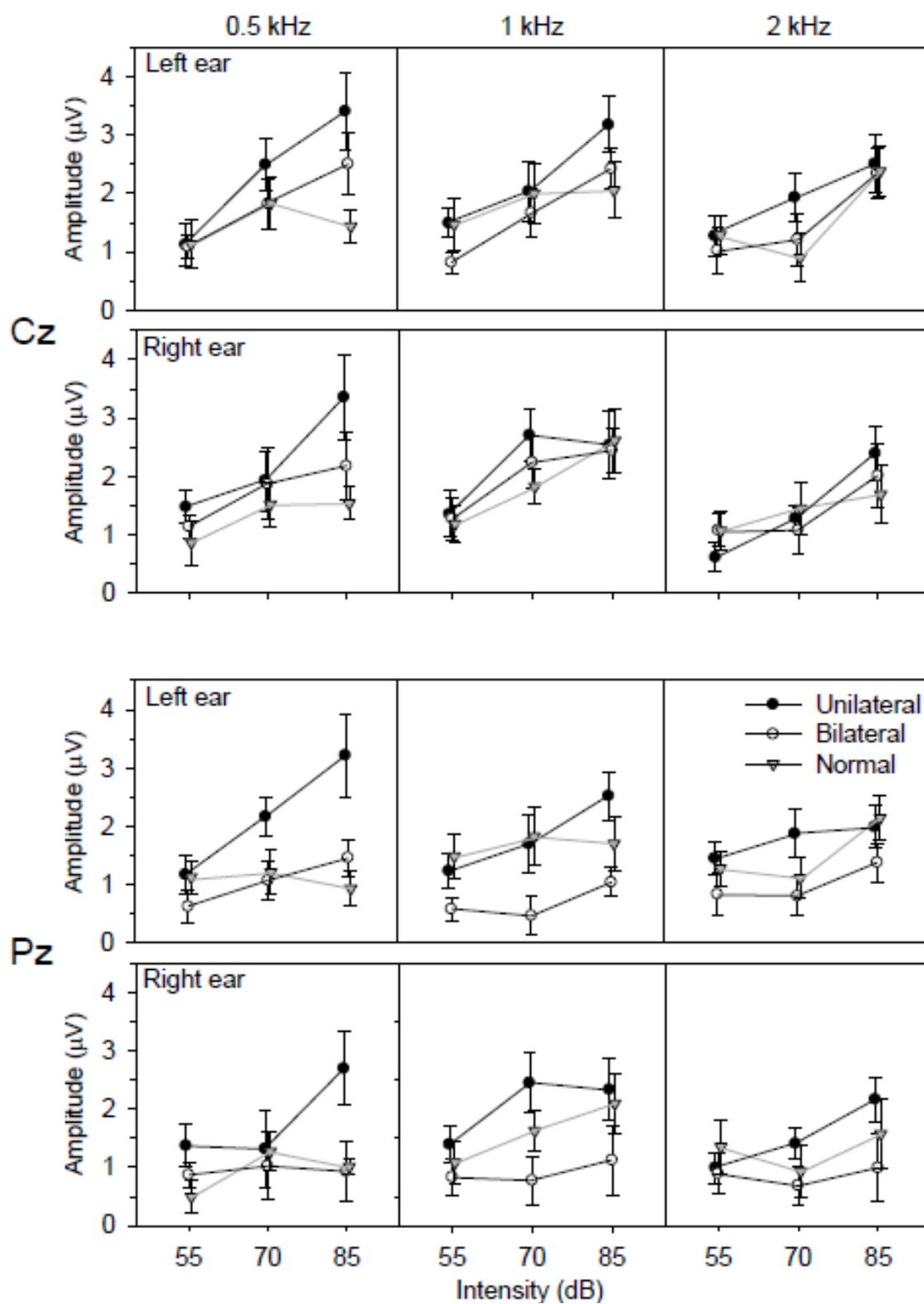
776

777 Figure 3



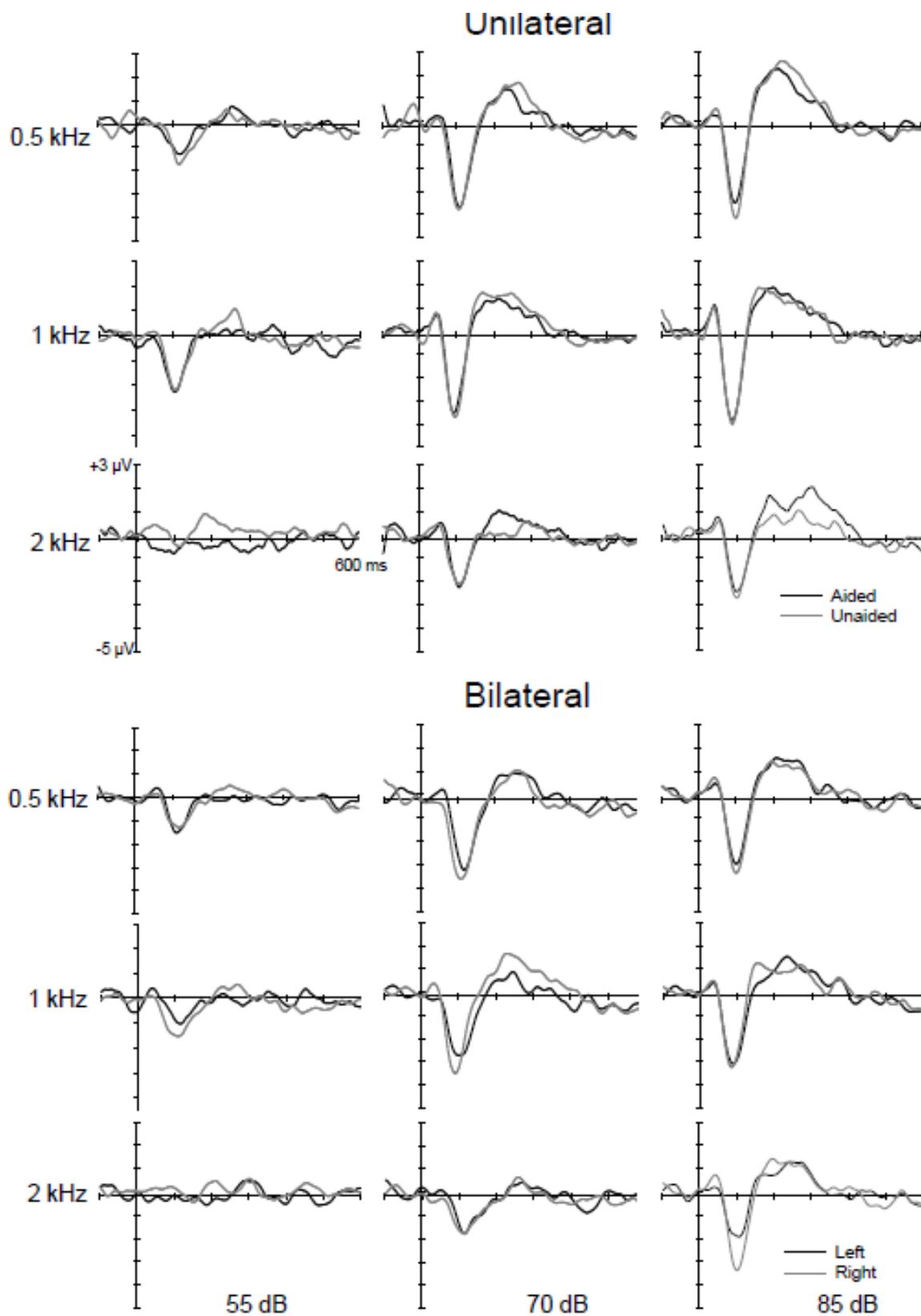
778

779 Figure 4



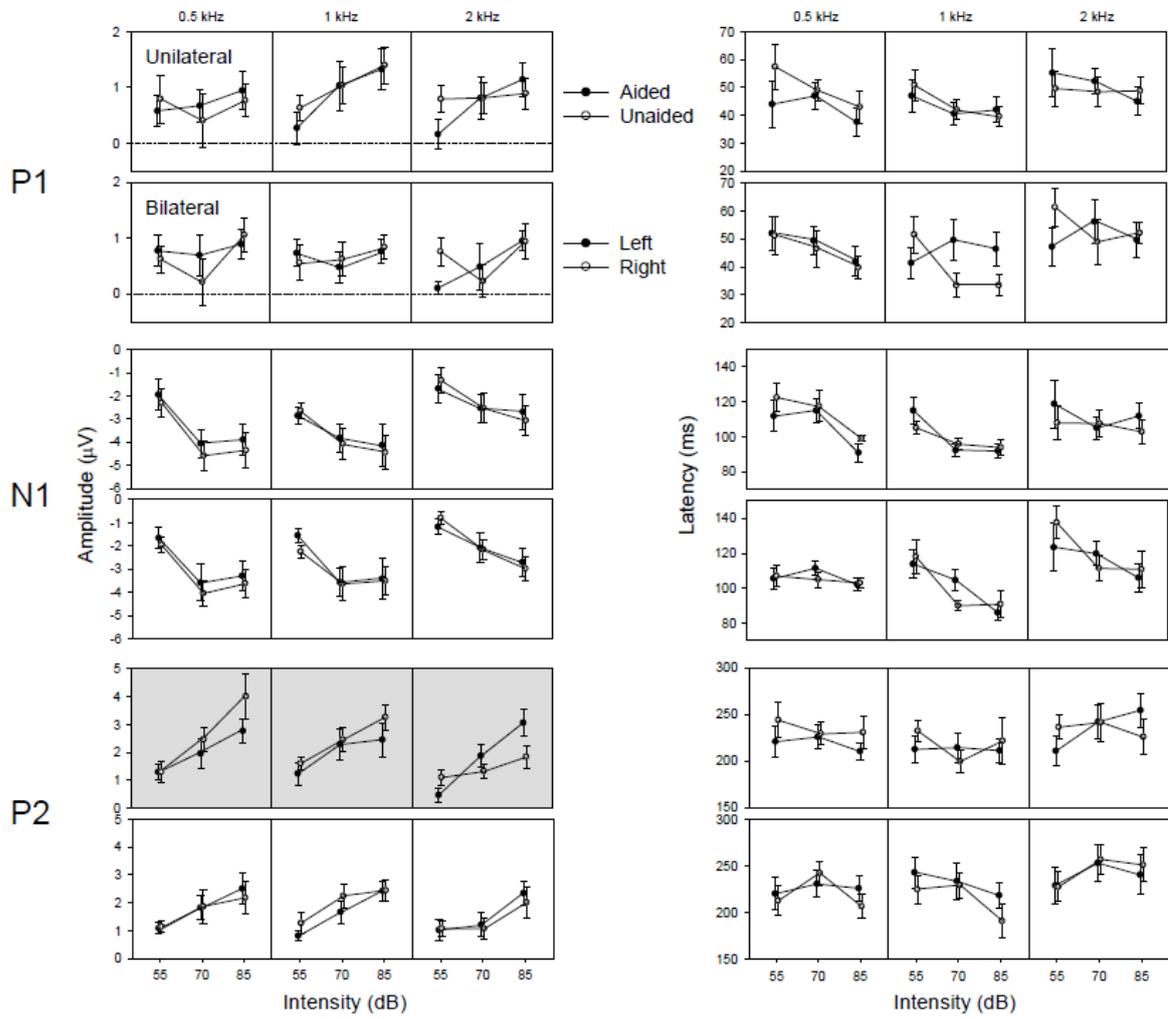
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781 Figure 5



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783 Figure 6



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Table 1: Results of speech audiometric tests. The ears of the normal and bilateral groups were grouped as left/right and the ears of the unilateral group as aided/unaided.

| Subject group | 50% speech discrimination (dB SPL) | | S/N ratio SPIN-test (dB SPL) | | Noise level SPIN-test (dB SPL) | |
|---------------|------------------------------------|-------------------|------------------------------|-----------------|--------------------------------|-------------------|
| | Right/unaided ear | Left/aided ear | Right/unaided ear | Left/aided ear | Right/unaided ear | Left/aided ear |
| Normal | 19.0 (SD 4.8) | 21.4 (SD 5.1) | 1.5 (SD 2.2) | 1.8 (SD 1.0) | 60.0 | 60.0 |
| Bilateral | 52.9 (SD 9.4) | 53.9 (SD 9.9) | 7.8 (SD 3.4) | 7.2 (SD 3.2) | 82.0 (SD 9.8) | 83.0 (SD 10.3) |
| Unilateral | 52.6 (SD 12.3) | 56.7 (SD 10.1) | 10.5 (SD 4.9) | 7.6 (SD 4.8) | 84.5 (SD 10.7) | 84.0 (SD 8.8) |

Table 2: F-values of repeated-measures ANOVAs for the AEP amplitudes and latencies at electrode sites Cz and Pz

| Factor | df | Amplitudes | | | | | Latencies | | |
|-------------------|-------|------------|----------|----------|---------------------|--------------------|-----------|----------|-------|
| | | P1 | N1 | P2 | P2 _{early} | P2 _{late} | P1 | N1 | P2 |
| Electrode site Cz | | | | | | | | | |
| Group (G) | 2, 27 | 1.73 | 0.56 | 1.14 | 1.15 | 0.36 | 5.04* | 3.59 | 1.05 |
| Ear (E) | 1, 27 | 0.00 | 3.19 | 0.69 | 2.20 | 0.00 | 0.16 | 0.05 | 1.67 |
| Frequency (F) | 2, 54 | 0.97 | 29.25*** | 6.27** | 10.58*** | 2.56 | 11.96*** | 18.99*** | 3.69 |
| Level (L) | 2, 54 | 7.83** | 20.51*** | 31.22*** | 23.60*** | 17.77*** | 0.57 | 17.18*** | 1.12 |
| G x E | 2, 27 | 0.22 | 2.43 | 0.55 | 1.04 | 0.88 | 0.42 | 0.76 | 3.07 |
| G x F | 4, 54 | 0.34 | 1.16 | 1.58 | 0.32 | 0.45 | 0.21 | 1.51 | 0.33 |
| G x L | 4, 54 | 1.80 | 2.04 | 1.36 | 1.49 | 0.98 | 0.94 | 0.95 | 0.47 |
| G x E x F | 4, 54 | 0.36 | 0.72 | 0.37 | 0.79 | 0.30 | 2.71 | 0.52 | 0.72 |
| Electrode site Pz | | | | | | | | | |
| Group (G) | 2, 27 | 0.66 | 0.57 | 5.43* | 5.80** | 2.17 | 0.19 | 2.07 | 0.81 |
| Ear (E) | 1, 27 | 0.02 | 0.58 | 0.92 | 1.96 | 1.40 | 0.57 | 0.03 | 0.00 |
| Frequency (F) | 2, 54 | 0.80 | 19.76*** | 0.81 | 2.31 | 1.31 | 4.40* | 9.28** | 0.53 |
| Level (L) | 2, 54 | 3.04 | 6.88** | 13.81*** | 5.09* | 4.55* | 1.01 | 0.02 | 4.14* |
| G x E | 2, 27 | 1.05 | 1.06 | 0.18 | 0.33 | 0.30 | 0.06 | 0.33 | 1.23 |
| G x F | 4, 54 | 0.52 | 0.91 | 2.99 | 1.37 | 0.87 | 0.68 | 0.51 | 0.58 |
| G x L | 4, 54 | 0.48 | 1.06 | 2.15 | 2.96 | 1.10 | 0.83 | 1.39 | 0.60 |
| G x E x F | 4, 54 | 1.00 | 0.14 | 0.23 | 0.56 | 0.27 | 1.52 | 1.38 | 0.60 |

* $p < 0.025$; ** $p < 0.01$; *** $p < 0.001$

Table 3: F-values of repeated-measures ANOVAs for the difference values of AEP amplitudes and latencies (unilaterally and bilaterally fitted group)

| Factor | df | Amplitudes | | | | | Latencies | | |
|-------------------|-------|------------|------|-------|---------------------|--------------------|-----------|------|------|
| | | P1 | N1 | P2 | P2 _{early} | P2 _{late} | P1 | N1 | P2 |
| Electrode site Cz | | | | | | | | | |
| Group (G) | 1, 18 | 0.10 | 0.01 | 0.60 | 0.17 | 0.05 | 1.02 | 0.00 | 1.66 |
| Frequency (F) | 2, 36 | 1.14 | 1.11 | 5.22* | 1.83 | 2.60 | 0.66 | 0.37 | 0.67 |
| Level (L) | 2, 36 | 1.42 | 0.32 | 0.43 | 0.29 | 1.96 | 1.91 | 0.60 | 0.66 |
| G x F | 2, 36 | 0.07 | 0.11 | 2.27 | 0.91 | 0.29 | 1.38 | 1.22 | 1.25 |
| G x L | 2, 36 | 0.51 | 0.27 | 0.71 | 0.68 | 0.03 | 0.79 | 2.22 | 1.62 |
| Electrode site Pz | | | | | | | | | |
| Group (G) | 1, 18 | 1.28 | 0.03 | 2.55 | 1.05 | 0.91 | 1.69 | 1.92 | 0.02 |
| Frequency (F) | 2, 36 | 0.07 | 1.25 | 3.58 | 1.72 | 1.60 | 0.47 | 1.14 | 0.06 |
| Level (L) | 2, 36 | 0.72 | 0.15 | 0.11 | 0.11 | 1.03 | 0.47 | 0.30 | 0.26 |
| G x F | 2, 36 | 0.78 | 1.29 | 2.08 | 0.79 | 0.85 | 0.98 | 0.45 | 1.77 |
| G x L | 2, 36 | 0.01 | 0.26 | 1.57 | 1.28 | 0.39 | 1.20 | 0.89 | 0.30 |

* $p < 0.025$

Table 4: F-values of repeated-measures ANOVAs for N1, P2, composite N1-P2 peak amplitudes and P2 mean voltages at electrode sites Cz and Pz (unilaterally fitted group)

| Factor | df | N1 | P2 | N1-P2 | P2 _{early} | P2 _{late} |
|-------------------|-------|---------|----------|----------|---------------------|--------------------|
| Electrode site Cz | | | | | | |
| Ear (E) | 1, 9 | 1.35 | 2.21 | 3.84 | 1.40 | 0.22 |
| Level (L) | 2, 18 | 8.93** | 17.67*** | 23.36*** | 12.36** | 12.57*** |
| Frequency (F) | 2, 18 | 11.32** | 4.42 | 15.73*** | 3.82 | 2.24 |
| E x L | 2, 18 | 0.37 | 0.32 | 0.33 | 0.20 | 0.90 |
| E x F | 2, 18 | 0.45 | 6.1* | 2.94 | 1.19 | 1.72 |
| Electrode site Pz | | | | | | |
| Ear (E) | 1, 9 | 0.39 | 4.84 | 3.09 | 1.20 | 0.26 |
| Level (L) | 2, 18 | 3.08 | 9.45** | 8.83** | 5.88* | 8.61** |
| Frequency (F) | 2, 18 | 5.84* | 1.48 | 13.8** | 2.06 | 0.51 |
| E x L | 2, 18 | 0.24 | 0.81 | 1.01 | 0.62 | 0.05 |
| E x F | 2, 18 | 1.26 | 3.97 | 5.55* | 1.29 | 1.36 |

* $p < 0.025$; ** $p < 0.01$; *** $p < 0.001$