Test Retest Reliability of Speech Evoked P300

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Abstract

The present study was taken with the aim to assess test retest reliability of speech evoked P300.The main objectives of the study were to access intra session and inter session test-retest reliability of P300 evoked by speech stimuli. 18 normal hearing individuals in the age range of 18 to 30 years participated in the study. Basic audiological evaluations such as pure tone audiometry, speech audiometry and Immitance evaluation were carried out using standard clinical procedures. P300 response was elicited using /da/(frequent) and /ba/ (infrequent) stimuli. To check the intra session reliability recording was repeated after 20 minutes without changing the position of the electrodes. To assess the inter session reliability recording was repeated after two days. Results showed no significant difference in amplitude and latency of P300 across three recording sessions indicating good reliability of P300. Cronbach's alpha revealed moderate to good reliability for P300 latency and amplitude respectively. Reliability estimates were better for intra session recordings compared to inter session recordings.

Key words: *Reliability, contralateral inhibition, otoacoustic emissions*

Introduction

P300 is an event related cognitive potential which is widely used to assess neural correlates of cognition and working memory. P300 was first described by Sutton et al.,(1965) and is evoked by active oddball paradigm. The most frequently used paradigm in P300 is two stimuli odd ball paradigm in which non frequent stimuli presented in series of frequent stimuli. Frequent stimuli will serve as standard stimuli and infrequent stimuli will be the target stimuli.Generation sites of P300 are complex with multiple sites that appear to be activated simultaneously. Generation sites includes frontal cortex (Courchesne, 1978) auditory cortex (Kileny, Robertson, 1985)and hippocampus and associated brain structures(Okada, Kaufman, &Williamson, 1983).P300 can be recorded in normal subjects as early as 250ms or as late as 400ms depending on the stimuli(Hall, 2007). Amplitude of P300 is more atCentro-parietal areas at the midline compared to other electrode locations.

Any sensory modality can be used to elicit P300 response in the descending order of clinical use these are auditory, visual, somatosensory, olfactory and gustatory (Bennington & Polich, 1999).In auditory modality tones or speech sounds can be used to elicit the responses. Auditory P300 is widely used to assess cognitive processing, hemispheric asymmetries, dichotic deficits, cerebral dysfunction, auditory perception skills and evaluation of language skills and efficiency. It has also been used in many clinical conditions to assess cognitive function. Demented patients was found to have more P300 latency than normal(Neshige, Barrett, & Shibasaki, 1988). Hemispheric asymmetries were found to be present in autistic children(Dawson, Finley, Phillips, Galpert, & Lewy, 1988). P300 can be used to find the severity of processing disorder in children with Central auditory processing disorders(Jirsa & Clontz, 1990). Study done by Guruprasad(2000) found that P300 can be used as a clinical tool in order to figure out learning disability in children. Autistic children was found to have decreased amplitude of P300(Niwa, Ohta, & Yamazaki, 1983).

The intrasubject consistency can be referred to as "the basic reliability of the response itself" (Segalowitz & Barnes, 1993)and is a fundamental indicator of the extent to which P300 response can be considered as a valid index of cognitive function, such as memory and information processing

In literature, a few studies have assessed the repeatability of P300 using tones or in other sensory modalities. Kinoshita et al.,(1995) assessed the reliability of P300 by repeating the recoding 8 times, within 7-10 days interval using two stimuli tones in oddball paradigm and the results revealed no significant difference between the waveforms. Sklare and Lynn (1984) assessed the test-retest reliability of latency and amplitude of P300 by using two stimuli oddball paradigm with tones. They investigated immediate and short term reliability on healthy adults in different time intervals and found that N1, P2, N2, P3 latencies and amplitudes were stable even after three days. Walhovd and Fjell (2002) assessed the one year test retest reliability of P300 using tonal stimuli in young and old adults and results revealed good reliability. Amplitude measures seemed more reliable compared to latency measures.

P300 is one of the widely used event related potentials to assess cognitive processing. It has been extensively used by audiologists and related professionals due to its varied clinical applications and to investigate finer aspects of auditory and cognitive-linguistic processing both in neuro-typical as well as in atypical individuals. However, to effectively use P300 as a clinical measure, it is very important to document the normal variations when the recording is repeated. There is scarcity of evidences regarding test-retest reliability of P300, especially speech stimuli. Hence for efficacious

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application of P300 reliability should be verified. Therefore the present study aimed to investigate the test retest reliability of P300 evoked by speech stimuli.

Methods

Participants

Twenty young adults in the age range of 18 to30 years(mean age= 24.8) participated in the study. All the participants had air conduction hearing thresholds within 15 dB HL at octave frequencies between 250 Hz to 8000 Hz. All participants had 'A'typetympanogram (Jerger, 1970; Lidén, 1969)and both ipsilateral and contralateral acoustic reflex thresholds were within 90 dB HL at 500 Hz and 1000 Hz. None of the participants reported exposure to loud noise, usage of ototoxic drugs, presence or history of ear discharge. All participants were right handed individuals(Oldfield, 1971) and passed screening test on auditory processing disorder(Keith, 1994)and Mini Mental Status Examination(Folstein, Folstein, & McHugh, 1975)

Equipment

Instruments used in the study were:

1. A calibrated audiometer GSI-61 with TDH 49 Earphones with MX-41/AR ear cushions for threshold estimation.
2. A calibrated GSI tympstarimmitance meter for evaluating middle ear status.
3. An Intelligent Hearing System (IHS) AEP system with smart EP for recording and analyzing P300.
4. A computer with Adobe Audition (Version 3) for recording and editing of the auditory stimulus.

Test environment

All tests were carried out in a sound treated room with noise levels within the permissible limits (American National Standards Institute, 2008)

Procedure

Basic audiological evaluation such as pure tone audiometry, speech audiometry and immittance evaluation were carried out using standard clinical procedures. Informed consent was taken from all the participants before the actual testing and participants were informed in prior regarding the details of the testing.

Electrophysiological testing

Stimuli

Two speech sounds /da/ and /ba/ of a native male Kannada speaker was recorded using Adobe Audition (Version 3). Figure 3.1 (a) and (b) shows the spectrogram and waveform of the stimuli used in the study.

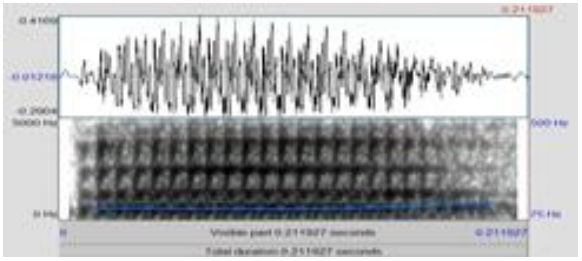

Figure 3.1 (a):Spectrogram and waveform of the stimulus /ba/

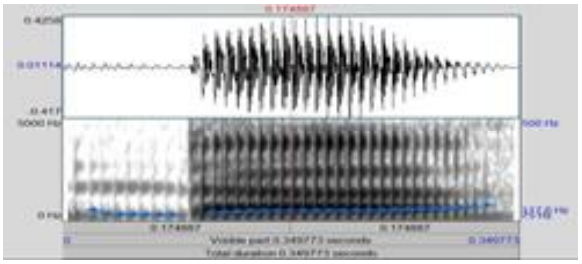

Figure 3.1 (b): Spectrogram and waveform of the stimulus /da/

Table 3.1: Protocol for P300 recording

Participant preparation

EEG was recorded by placing the electrodes on frontal (Fz), vertex (Cz) and parietal (Pz)sites with nasion as reference. Ground electrode was placed on the right mastoid. Electrooccuogram was recorded with electrodes placed above and below right eye. Impedances of all electrodes were kept at or below 3 k?. To ensure the correct and consistent positioning of the electrodes following procedure was followed: distance between nasion (bridge of the nose) to inion (occipital protrusion) and distance between both zygomatic notches were measured and the total length was noted in centimeters. The midpoint of these distances - located at 50% of the total length between nasion to inion and between both zygomatic notches was marked as the point Cz (vertex point).20% of the total distance from nasion to inion towards front of Cz along the midline was marked as Fz and towards back of Cz along midline was marked as Pz.

Table 3.1 shows the test protocol for P300 recording.

Type of stimulus	Speech sounds (frequent: /da/and infrequent:/ba/)
Polarity	Rare fraction
Filters	1-30Hz
Intensity	80 dBnHL
Number of Stimulus	200 (80% frequent stimulus and 20% infrequent stimulus)
Transducer	ER-3A
Repetition Rate	0.8/s
Presentation	Binuaral
Electrode Montage	Inverted: Nasion Non Inverted: Fz,Cz,Pz Ground :Mastoid

Test Procedure

Participants were explained about the testing procedure and also the tasks to be performed. The participant was then seated comfortably and electrodes were placed. Participants were then instructed to:

- Relax and remain alert throughout the testing.
- Keep their eyes open and to fixate their vision to one spot.

They were asked to make a mental count of infrequent stimulus (ba) in series of frequent stimulus (da).At the end of the recording they were asked to report the number of stimuli counted. This was done in order to make sure that the subjects give attention to the stimuli and these results were not used for any other purpose.

ER-3A inserts were placed in subject's ears and the stimuli were presented binaurally through the inserts. Waveforms of both frequent and infrequent stimulus response were recorded from all the three electrode sites. Electroocculogram was recorded with electrodes placed above and below right eye. Artifact rejection at ocular channel was adjusted to reject all eye blinks. Sweeps affected by eye-blinks were automatically rejected across all channels by the ocular channel artifact rejection criterion. LLR was recorded for the deviant/ infrequent stimuli using the same protocol that was used to record the P300 response from all the three electrode sites(Fz,Cz,andPz). Following the first recording, 20 minutes' rest time was given to participants and after that P300 was recorded again without altering the position of the electrodes. Electrode impedances were ensured to be the same as first P300 recording.Following second recording participant was released and third P300 was recorded 2 days later using the same protocol and procedure described above. All basic audiological evaluations were repeated prior to P300 testing. Through a structured interview, it was ensured that there were no significant auditory, cognitive or neurological problems between the recordings.

Results

The aim of this study was to investigate the test-retest reliability of P300 evoked by speech stimuli in individuals with normal hearing. For this purpose, P300 was recorded thrice and was compared across recordings sessions to determine intra and inter session reliability. Data obtained was analyzed at two levels - at group and individual level.

Characterization of P300

Figure 4.1 shows grand averaged waveform for deviant stimuli and LLR across three electrode sites and three recording sessions. From the Figure 4.1, it can be inferred that deviant stimuli waveform (red) had large positivity between 230 ms to 320 ms which is present in LLR (black). This confirms the presence of P300. From the Figure 4.1, it can also be seen thatPz electrode had higher P300 amplitude followed by Cz and then Fz.

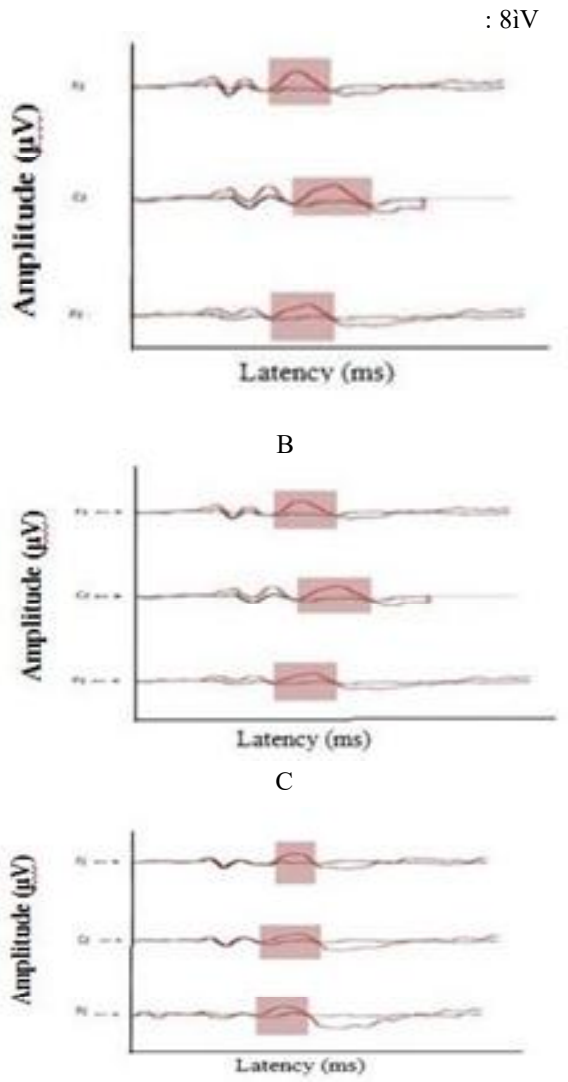

Figure 4.1: Grand averaged waveform for deviant stimuli and LLR across three electrode sites for a) recording 1 b) recording 2 c) recording 3. The each figure first tracing from Fz site, second from Cz and third from Pz site.

Group data Analysis

Repeatability of P300 was measured by assessing waveform modulation, peak latency and amplitude.

Waveform modulation

Statistical significance of the differences between P300 wave forms recorded in intra-session (recording 1 vs. recording 2) and inter-session (recording 1 vs. recording 3) was assessed by carrying out significance tests at every time point using randomization procedure. Figure 4.2 shows grand averaged waveforms recorded in intra-

session across different electrode sites. Spikes in the green bars below the waveforms represent the time regions where two waveforms differed from each other significantly ($p < 0.05$). From the Figure 4.2, it can be seen that there was no statistically significant difference between two recording sessions in all three electrode sites.Figure 4.3 shows grand averaged P300 waveforms obtained in recording 1 and 3 at three electrode sites. Spikes in the green bars below waveforms represent the time regions where two waveforms differed from each other significantly ($p<0.05$). From the Figure 4.3, it can be seen that there was no statistically significant difference between two recording sessions in all three electrode sites.

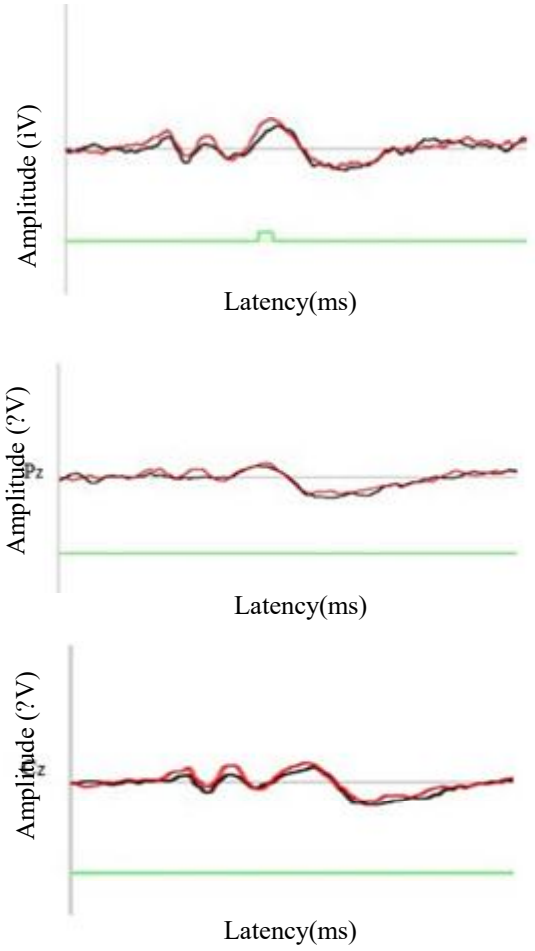

Fig 4.2: Grand averaged P300 waveforms obtained in recording 1 and 2 at three electrode sites X axis is time in ms and Y axis is amplitude in uV.

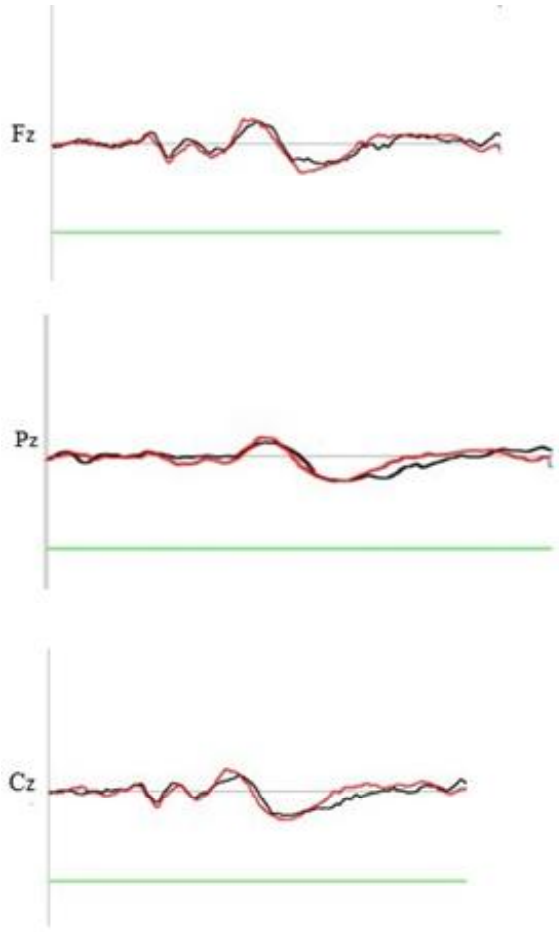

Fig 4.3: Grand averaged P300 waveforms obtained in recording 1 and 3 at three electrode sites. X axis is time in ms and Y axis is amplitude in uV.

Peak Latency and amplitude

In the grand average P300 waveform onset and offset latency of P300 was noted for each electrode and recording sessions separately. In individual waveforms the region that had maximum amplitude between onset and offset latency of grand averaged P300was considered as P300 and its peak amplitude and latency was noted for further statistical analyses.Figure 4.4 shows the mean and the standard deviation of P300 latency in three recordings. From the Figure 4.4, it can be seen that latency of P300 did not vary much between the recordings. To assess the statistical significance of differences in latency across recordings, a repeated measure ANOVA was performed. Repeated measures ANOVA did not reveal a significant main effect of recording condition [$F (1.9, 32) = 0.66, p<0.05$]. However, there was significant main effect of electrodes on peak latency [$F (1.8, 34)= 33.8, p <0.05$]. Interaction between electrodes and latency was not significant. Chronbach'salpha was calculated as reliability estimates (only for Pz electrode as it had higher amplitude). Chronbach's alpha was 0.6 for intra session and 0.5 for inter session recording of P300 latency.

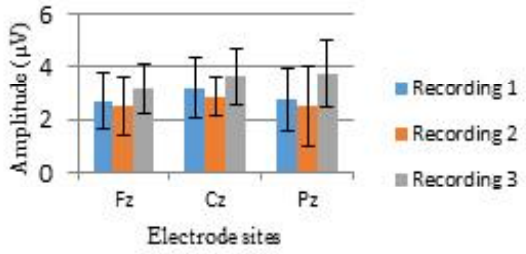

Figure 4.4: Mean and the one standard deviation of P300 latency in three recordings.

Figure 4.5 shows the mean and one standard deviation of P300 amplitude in three recordings. From Figure 4.5, it can be seen that amplitude of P300 did not vary much between the recordings. To assess the statistical significance of amplitude differences across recordings, a repeated measure ANOVA was performed. Repeated measures ANOVA did not reveal a significant main effect of recording condition[$F(1.3, 23.1)=0.54$, $p<0.05$] and electrodes [$F(1.2, 21.7) =3.5$, $p<0.05$]on amplitude.Interaction between electrodes and amplitude was also not significant. Chronbach's alpha was 0.7 for intra session and 0.5 for inter session recording of P300 amplitude.

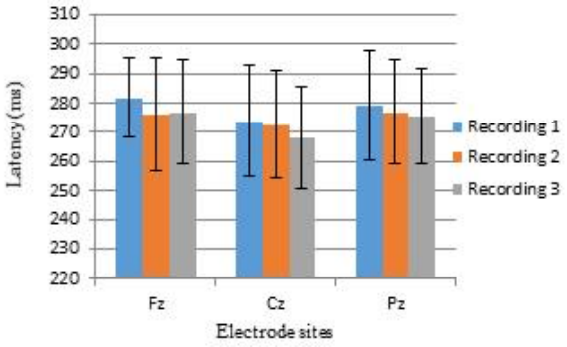

Figure 4.5: Mean and one standard deviation of P300 amplitude in three recordings.

The present study aimed at assessing the intra and inter session reliability of speech evoked P300. Results indicated that P300 latency had moderate intra and inter session reliability and amplitude had good intra inter session reliability. To our knowledge, this is the first study to evaluate the intra and inter session reliability of speech evoked P300. Previous studies have measured reliability of tone evoked P300 and found reliability estimates similar to current study(Sklare& Lynn, 1984). Segalowitz and Barnes (1993) measure reliability of tone evoked P300 on 19 adolescents across 1 year 10 months interval. They assessed within session as well as across session reliability of P300 amplitude and latency. Their results indicated that P300 had good within session and across session stability both for amplitude and latency. P300 latencies were slightly more stable compared to amplitudes. Katayama and Polich (1996)measured reliability of tone evoked P300 on 100 undergraduate students and concluded that P300 amplitude and latency was highly reliable. Similarly Hong et al.(2013) also reported good intra session reliability of tone evoked P300. They measured P300 in an odd ball paradigm for 1000 Hz standard and 2000 Hz deviant tones on 30 normal hearing adults. P300 latency had excellent reliability and whereas reliability of the amplitude was fair to good.Hall (2007) assessed the test retest reliability amplitude and latency of P300 in 19 monozygotic twins twice at the interval of 7 and 56 days. Their results showed very high reliability estimates for both amplitude and latency of P300.

Consistent with the previous report even the current study found high reliability of P300. Reliability estimates were higher for amplitude compared to latency. Reliability estimates were higher in intra session recordings compared to intersession recordings. Our results suggest that P300 is a reliable measure and has necessary stability required for group research. Reliability estimates are not satisfactory for individual applications. Among the parameters, amplitude had higher reliability estimates and is a more stable measure for individual application. However, it is advised to interpret P300 parameters for clinical purpose with caution.

Conclusions

These results suggest that P300 is a reliable measure and has necessary stability required for group research. Reliability estimates are not satisfactory for individual clinical applications. Among the parameters amplitude had higher reliability estimates and is a more stable measure for individual application. However, it is advised to interpret P300 parameters for clinical purpose with caution.

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