ARTICLE Quantitative and Qualitative Representation of Introductory and Advanced EEG Concepts: An Exploration of Different EEG Setups

Shelby L. Hatton¹, Shubham Rathore², Ilya Vilinsky^{1, 2}, Annette Stowasser^{1,2}

¹Undergraduate Neuroscience Program, University of Cincinnati, Cincinnati, OH 45221; ²Biology Department, University Of Cincinnati, Cincinnati, OH 45221.

https://doi.org/10.59390/GEBE4090

Electroencephalograms (EEGs) are the gold standard test used in the medical field to diagnose epilepsy and aid in the diagnosis of many other neurological and mental disorders. Growing in popularity in terms of nonmedical applications, the EEG is also used in research, neurofeedback, and braincomputer interface, making it increasingly relevant to student learning. Recent innovations have made EEG setups more accessible and affordable, thus allowing their integration into neuroscience educational settings. Introducing students to EEGs, however, can be daunting due to intricate setup protocols, individual variation, and potentially expensive equipment. This paper aims to provide guidance for introducing students and educators to fundamental beginning and advanced level EEG concepts. Specifically, this paper tested the potential of three different setups, with varying channel number and wired or wireless connectivity, for introducing students to gualitative and quantitative exploration of alpha enhancement when eyes are closed, and observation of the alpha/beta anterior to

INTRODUCTION

Electroencephalography (EEG) is a non-invasive technique that records the electrical activity of cortical brain regions using electrodes placed on the scalp (Britton et al., 2016). Incorporating the EEG technique into an undergraduate teaching laboratory is of interest for educators because this technique is widely used in research (Bazzani et al., 2020), in medicine for the diagnosis of epilepsy (Noachtar et al., 2009), and for the diagnosis of other neurological dysfunctions (Rivera et al., 2022). Moreover, the EEG technique has become increasingly popular because of its applicability for brain-computer interface (BCI) (Sahar at al., 2021), neurofeedback (Demos, 2019), and investigating evoked potentials/event related potentials (Miller et al. 2008). Thus, introducing EEG techniques allows undergraduate students to explore human brain activity and obtain skills for real world applications. We tested three EEG setups for their potential to detect fundamental EEG activity patterns that are essential in clinical and research settings. Specifically, an undergraduate student recorded, visualized, and performed quantitative analysis of alpha and beta waves in seven fellow students. The ultimate goal was to provide guidance for facilitating a holistic education in EEG recording. This includes both the qualitative visual inspection, on an individual basis, which is utilized more in clinical diagnostic settings, and the quantitative EEG analysis utilized in the investigative research fields.

posterior gradient. The setups were compared to determine their relative advantages and their robustness in detecting these well-established parameters. The basic 1- or 2channel setups are sufficient for observing alpha and beta waves, while more advanced systems containing 8 or 16 channels are required for consistent observation of an anterior-posterior gradient. In terms of localization, the 16channel setup, in principle, was more adept. The 8-channel setup, however, was more effective than the 16-channel setup with regards to displaying the anterior to posterior gradient. Thus, an 8-channel setup is sufficient in an education setting to display these known trends. Modification of the 16-channel setup may provide a better observation of the anterior to posterior gradient.

Key words: Electroencephalogram (EEG); Power Spectrum Analysis; Alpha:Beta Power Ratio; Eyes-Closed Test

EEGs capture the sum of both excitatory and inhibitory electrical postsynaptic potentials from large numbers of neurons. Synchronous firing activity of neurons in cortical regions leads to what is called "brainwaves" (Kirschstein et al., 2009). These basic principles of the EEG were defined by its developer Hans Berger who visualized the first human brainwaves in 1924 (Ince et al., 2021) and was the first to coin the terms "alpha" and "beta" waves based on their frequency (Berger, 1929). It was found that beta waves (13 - 35 Hz) were more dominant in the fronto-central area. On the other hand, Alpha waves (8 - 13 Hz) were more dominant in the posterior regions and thus were termed as the posterior dominant rhythm (Britton et al., 2016). The higher frequency beta activity in the anterior fronto-central region and lower frequency alpha activity in the posterior occipital region later became known as the anterior to posterior gradient (Figure 1).

As the origin of waveforms were discovered, their reactivity was investigated as well. Alpha waveforms were found to react to eyes-open and eyes-closed states with alpha attenuation during eyes-open and alpha accentuation during eyes-closed (Figure 2). This phenomenon became a foundational concept known for EEGs (Britton et al., 2016). These two concepts have become "gold standard" testing practices in the routine neurodiagnostic EEG as set per the American Clinical Neurophysiology Society (ACNS) guidelines (Tatum et al., 2016). These two trends are among

Hatton et al. Quantitative and Qualitative Representation of Introductory and Advanced EEG Concepts Laboratory A143





the most visualized aspects of the EEG signal and are important in the education of undergraduate neuroscience students (Britton et al., 2016).

More advanced methods for the students to learn includes quantitative data analysis. Quantitative EEG (QEEG) is an analysis technique that utilizes complex signal processing to determine numerical representations of the EEG (Fetz, 2007). QEEG is becoming more widely used not only in the diagnosis of many disorders (Kopańska et al., 2022) but also in research (Bazzani et al., 2020), BCI (Wange et al., 2019), and neurofeedback (Demos, 2019). One commonly used fundamental technique in QEEG is power spectrum analysis. This breaks the EEG into its frequency components based on the power of the signal. Power is defined as the amplitude of the frequency component squared and can be utilized to calculate power ratios (Dempster, 2001). These ratios can fundamentally be applied to the previously mentioned basic concepts. Alpha:beta power ratios are expected to be highest with eyes closed and in the posterior regions due to larger alpha power but lower beta power in these regions/states. This understanding of alpha in respect to beta is a valuable next step for students to explore after introductory qualitative evaluation.

Hands-on setup of the EEG is imperative to student learning (Segawa, 2019). This includes the consideration of the environment to lessen electrical noise and artifacts, electrical grounding, cost effectiveness of EEG equipment, handling of the equipment, and overall design of the experiment to ensure quality results (Ledwidge et al., 2018). When choosing equipment, one must consider the number of channels and compatible electrode positions for student learning. In general, EEG equipment and setups can range anywhere from one channel (Shields et al., 2016) to 256 channels (Feng et al., 2016). In this study, setups of one, two, eight, and 16 channels were utilized with different



Figure 2. Spectrogram of frequency bands during eyes-closed and eyes-opened states from subject 4. Yellow indicates higher power while red indicates lower power. The more prominent yellow "band" located near 10 Hz indicates the higher power of alpha during the eyes-closed state. This yellow "band" attenuates in the eyes-open state, thus indicated by the orange and red bands.

electrode positions. For each setup, we explored the potential for introductory and advanced level exercises. More specifically, we determined the ability of the setup to detect the anterior to posterior gradient and alpha attenuation to eyes-open state.

MATERIALS AND METHODS

Subjects

Subjects were volunteers who were currently enrolled at the University of Cincinnati Main Campus in March and April of the 2022 spring semester. Subjects included 3 males and 4 females for a total of 7 subjects. Subjects were within the age range of 20-22 years old. Subjects were asked to maintain their normal routines and eating habits for the day of the experiment to replicate the setting of a normal day in the classroom. All experiments on subjects were carried out by an undergraduate student attending the neuroscience teaching lab.

EEG Hardware and Settings

The 1-channel and 2-channel setups utilized the PowerLab 26T from A/D instruments with a sampling rate of 1000Hz. The 8-channel setup utilized the Unicorn Hybrid Black from g.tec with a sampling rate of 250Hz. The 16-channel setup utilized Cyton+Daisy board and electrode cap from OpenBCI with a sampling rate of 128Hz. The 1 and 2 channel setup utilized gold plated electrodes and Ten20 conductive paste while the 8- and 16-channel setups utilized Ag/AgCI coated electrodes and conductive gel provided by g.tec. The specifications of the setups are summarized in Table 1.

Fundamental EEG Definitions

In EEG recordings and as seen in Table 1, many definitions and concepts are utilized when setting up the EEG hardware and settings that must be understood by the student. To start, students must make several decisions about how to *digitally acquire and display the data*. This includes selecting the sampling rate, montage, and filters. *Sampling rate* is defined as the amount of data points collected per second. This is typically expressed as frequency. Per ACNS

CHANNELS	1	2	8	16
MONTAGE:	Bipolar	Bipolar	Referential	Referential
PRODUCT:	26T Powerlab Bio amplifier	26T Powerlab Bio amplifier	Unicorn Hybrid Black	Cyton + Daisy Biosensing Board
MANUFACTURER:	AD Instruments	AD Instruments	g.tec	OpenBCI
GROUND:	Ear Lobes	Ear Lobes	Over the Mastoid Bones	AFZ
REFERENCE:	OZ	OZ, PZ	Over the Mastoid Bones	CPZ
ELECTRODE TYPE:	Wet	Wet	Wet	Wet
IMPEDANCE CHECK:	No	No	Yes	Yes
SAMPLING RATE:	1000Hz	1000Hz	250Hz	128Hz
INTERNAL PROCESSING:	No	No	Yes	Yes
BANDPASS FILTER:	None	None	1-30Hz	1-50Hz
60 HZ NOTCH:	Off	Off	On	On

Table 1. Summary of EEG hardware and settings specifications.

guidelines, a minimum sampling rate of 256 Hz must be used for diagnostic purposes (Tatum et al., 2016). However, many commercial and some research systems utilize less, like the 8- and 16-channel setup (Table 1) In this study, 250Hz and 128Hz were used based on the setting limitations in the EEG systems.

Next, a montage must be selected. Montages are displays of ordered arrangements of the recordings from different electrode positions. This is often defined by the standardized 10-20 system in which distance between electrode positions are either 10% or 20% of the total skull measurement from left-to-right or front-to-back (Archarya et al., 2019). An alternative system is the 10-10 system that utilizes interelectrode distances of 10% of the total measurements (Nuwer, 2018). EEGs utilize differential amplifiers that measure the voltage difference between two sites. Therefore, two electrodes are utilized. These electrodes are termed the active (primary or positive) electrode and the secondary (negative or reference) electrode. The primary electrode is placed above the desired location for recordings and measurements. The secondary electrode position acts as a reference to which the primary electrode's voltage is compared and from which it is subtracted. There are two types of montages that are defined based on the selection of the secondary electrode. One type is termed a *referential montage* and occurs when the secondary electrode for all channels is a single reference or commonly chosen electrode position (Kutluay, 2019). This reference tends to be an area that is electrically quiet such as the area over a bone or soft tissue. The region over the mastoid bone or earlobes are often utilized. Referential montages are better for displaying larger amplitude waves or abnormalities that show large distribution across the brain. The second group of montages are termed bipolar and occur when the secondary electrode chosen is a nearby or adjacent electrode. Choosing electrodes near each other allows a bipolar montage to show spatial differences between specific brain sites and better visualizes smaller amplitude waveforms or abnormalities (Kutluay, 2019). In this study, both montage types were utilized, depending on specifications of the EEG equipment. Another concept utilized in an EEG is the *ground electrode* which is typically utilized to reduce noise (electrical interference) from the recording by subtracting its potential from both the primary (active) electrode and secondary (reference) electrode (Luck, 2022).

A *filter* is utilized to remove or decrease preselected frequencies from the recording and allows the desired frequencies to be shown. One example of a filter is a notch filter. This filter is specialized for attenuating a specifically chosen frequency. An example is a *60Hz notch* filter that decreases the 60 Hz signal that is produced by electrical equipment in the United States. Another filter type is a *bandpass filter*. This filter allows the passing of a range of frequencies. All frequencies outside this range are attenuated by the filter, thus allowing for a window or bandwidth of frequencies to be more easily seen (Britton et al., 2016).

Finally, students should understand the importance of checking electrode *impedances*. While resistance is the measurement of opposition to a direct current, impedance is the measurement of opposition in an alternating current. Some impedance is needed to obtain a signal, but if an impedance is too high, less of the biological signal is allowed through, negatively affecting the signal to noise ratio. Most EEGs utilize impedance checks to ensure signal quality. The 1- and 2-channel setups did not check impedances due to an absence of this function in their equipment (Tatum et al., 2018).

Electrode Placement

The 1-channel setup utilized a bipolar montage. The primary electrode was placed at FPZ, according to the 10-20 system (Figure 3), and the secondary electrode was placed on OZ. The channel 1 (FPZ-OZ) setup provides single channel coverage of the cerebral cortex with the largest interelectrode distance, as is recommended by A/D instruments and Shields et al. (2016). A ground electrode was placed on the left ear lobe (A1). The 2-channel setup tested the importance of interelectrode distance by comparing channels that utilized two different distances between the electrodes. Like the 1-channel setup, it utilized a bipolar montage. Channel 1 was set up as before (FPZ-OZ). Channel 2 electrodes were placed closer together (FZ-PZ); more specifically, the primary electrode was placed at FZ and the secondary electrode at PZ, resulting in channel 1 having a larger interelectrode distance than channel 2

The 8-channel setup utilizes a referential montage and placements in both the 10-20 and 10-10 international system. The setup is as follows: channel 1: FZ-M1, channel 2: C3-M1, channel 3: CZ-M1, channel 4: C4-M1, channel 5: PZ-M1, channel 6: PO7-M1, channel 7: OZ-M1, and channel 8: PO8-M1. The reference (M1) and ground (M2) are positioned on the mastoids behind the ears. With this setup, we tested the potential benefit of utilizing a multi-channel system. In the standard configuration, this setup comes with a wireless amplifier which attaches to the electrode cap, and shorter leads to electrodes.

The 16-channel setup also utilizes a referential montage and placements in the 10-20 and 10-10 international system. The setup is as follows: channel 1: FP1-Ref,



Figure 3. Summary of electrode placement for all setups. Following the 10-20 and 10-10 system, FPZ and OZ are utilized in 1-channel setup and are colored green. FPZ, FZ, PZ, and Oz are utilized in the 2-channel setup and are outlined in yellow. Fz, C3, CZ, C4, PZ, PO7, OZ, and PO8 are utilized in the 8-channel setup and are outlined in red. FP1, FP2, F7, F3, FZ, F4, F8, C3, C4, T5, P3, PZ, P4, T6, O1, O2, FCZ, and CPZ are utilized in the 16-channel setup and is colored blue. Frontal-Polar (FP), Frontal (F), Central (C), Parietal (P), Temporal (T), Occipital (O), Zero (Z), Mastoid (M), Auricular (A).

channel 2: FP2-Ref, channel 3: F7-Ref, channel 4: F3-Ref, channel 5:FZ-Ref, channel 6: F4-Ref, channel 7: F8-Ref, channel 8: C3-Ref, channel 9: C4-Ref, channel 10: T3(P7)-Ref, channel 11: P3-Ref, channel 12: PZ-Ref, channel 13: P4-Ref, channel 14: T6(P8)-Ref, channel 15: O1-Ref, channel 16: O2-Ref. The ground electrode is placed at FCZ and the reference electrode is placed at CPZ (Figure 3). With this setup, we tested the benefit of utilizing more than 8 channels, as is used in clinical settings. In its standard configuration, just like the 8-channel setup, this setup comes with a wireless amplifier. The amplifier is not, however, attached to the electrode cap, and attaches to the electrodes with longer leads.

Experimental Protocol and Data Collection

For each setup, the subjects were given one minute to relax and acclimate. Then the subjects were asked to open their eyes and stare at a stationary object for 30 seconds. After 30 seconds of eyes opened, the subjects were asked to close their eyes for 30 seconds. This was performed a total of three times, resulting in six 30 second trials: three with eyes open and three with eyes closed. Care was taken to ensure consistent application of electrodes and conduction of the protocol to minimize statistical variance (Naydenov, 2022). First, the raw recording of each setup was visualized and evaluated to determine its potential for introductory level identification of brainwaves. This data was collected and visualized by an undergraduate student. The brainwaves were evaluated by having the student count the waves per second to determine the frequency. If the student was able to see alpha and beta wave frequencies in a stable recording, then it was considered to be an efficient visualization of brainwaves. For more advanced level education, power spectrum analysis was performed by transforming the signal into its frequency components and then obtaining power values for each subject during their 30 second trials. The power of alpha (8Hz-13Hz) and of beta (13Hz-30Hz) within these trials were calculated using LabChart 8 (ADInstruments) with built in data analysis tools. These values were used to create alpha:beta power ratios for each trial. After the ratios were obtained from the power spectrum analysis, the alpha:beta power ratios were averaged to create an overall alpha:beta ratio for eyes open and eyes closed. All raw EEG data was viewed, filtered, and analyzed via LabChart 8 (ADInstruments). Figures were made utilizing Excel, R, and photoshop.

Quantitative Definition of the Known Trends

Two foundational EEG patterns were explored in this experiment. The first is alpha reactivity to eyes-open and eyes-closed states. The second is the presence of the anterior to posterior gradient. Subjects were considered to follow the first pattern if their average alpha:beta ratios for all channels were higher when their eyes were closed vs. when open. For the second pattern, the alpha:beta power ratios of the anterior electrodes during the eyes-closed condition were averaged and compared to the average ratio of the subjects' posterior electrodes. If the posterior averaged ratios were higher than that of the anterior electrodes, then the subject was considered to follow that pattern. Statistical significance was then tested using the same parameters above. A paired, two-tailed, T-test was utilized with a critical p-value of p= 0.05.

RESULTS

When comparing the setups, first a qualitative assessment of the EEG recording was performed. The student examined the change in brainwave frequency and morphology when the eyes were opened vs. closed in all setups. They were able to count these frequencies and determine they saw primarily beta waves with eyes-open and primarily alpha waves with eyes-closed, thus indicating all setups were adept at visualizing brainwaves and identifying the patterns.

The 1-channel setup places electrodes at the posterior and anterior of the scalp and provides a whole cortex overview of alpha and beta waves. Due to the lack of multiple electrode distribution on the scalp, only the eyesopen and eyes-closed trend was considered. Four of the seven subjects (57%), subjects 3, 4, 5, and 7, showed the expected pattern of higher alpha:beta power ratios being seen when eyes were closed vs. open (Figure 4), whereas subjects 1, 2, and 6 showed the opposite pattern.

To determine if distance or origin of oscillations affected



Figure 4. A 1-channel EEG recording during eyes-open and eyesclosed. (*A*) Visualization of the raw EEG recording. (*B*) Alpha:beta power ratios of the 30 second eyes-open and closed trials. This setup utilized the positions FPZ-OZ. Higher alpha:beta ratios were expected when eyes were closed. Error bars were determined by calculating standard error within each subject for all trials. Solid bars indicate the eyes-open and striped bars indicate the eyesclosed conditions.

the whole brain recording in the 1-channel setup, a closer electrode distance was used in the 2-channel setup. Due to the lack of multiple electrode distribution on the scalp, only the eyes-open and eyes-closed pattern was considered. The 2-channel setup resulted in only two subjects out of seven (29%), subjects 3 and 5, following the expected pattern in all channels (Figure 5). Interestingly, some individuals had conflicting results in which one channel had the expected pattern and the other didn't. This could be due to the effect of interelectrode distance on the amplitude of waveforms. This contradiction was observed in four of the seven (58%) subjects: 1, 2, 5 and 7. Subject 6 showed opposite results than the expected pattern in both of their channels.

For the 8-channel setup (Figure 6), all subjects showed at least one expected pattern. Five out of seven (71%) subjects (2, 3, 4, 5, and 6), showed both expected patterns. Subjects 1 and 7 out of the seven subjects (29%) showed only one expected pattern. Subject 1 showed the expected pattern of higher alpha:beta ratios in the posterior head region than in the anterior head region but did not show the expected higher alpha:beta ratios when the eyes were closed instead of open. Subject 7 showed the expected pattern of higher alpha:beta ratios when the eyes were



Figure 5. A 2-channel EEG recording during eyes open and closed. (*A*) Visualization of the raw EEG recording. (*B*) Alpha:beta power ratios of the 30 second eyes-open and closed trials. The positions FPZ-OZ for channel one and FZ-PZ for channel two were utilized. Error bars were determined by calculating standard error within each subject for all trials. Solid bars indicate the eyes-open and striped bars indicate the eyes-closed condition.

closed instead of open, but they did not show the pattern of higher alpha:beta ratios in the posterior head region.

The 16-channel setup (Figure 7) was selected to replicate that of a clinical or research setting. This allowed comparisons of the other setups to what is considered the "gold standard" of the field. Four of seven (57%) subjects (3, 5, 6, and 7) showed both expected patterns (Figure 7). Two of seven (29%), subjects (2 and 4), showed one expected pattern. Subject 2 showed the anterior to posterior gradient but not the eyes closed pattern. Subject 4 showed higher alpha:beta ratios when eyes were closed, but they showed the reverse of the anterior to posterior gradient. One subject, Subject 1, of seven (14%) showed no expected pattern.

Utilizing power spectrum analysis allowed for a quantification of alpha and beta wave content instead of simply estimating their amount via qualitative inspection. When comparing the overall quantitative results in all channels (Figure 8), the 8-channel setup was the most reliable in detecting the expected trends with 100% showing the eyes-open and closed pattern and 86% showing the anterior to posterior gradient. The 16-channel setup that was closest to medical and research EEG setups was less reliable in quantitative detection of the known patterns with only 86% showing the eyes-open and closed pattern and 71% showing the anterior to posterior to posterior to posterior gradient. The 2-



Figure 6. An 8-channel EEG recording of eyes-open and closed trials. *(A)* Visualization of the processed recording with a bandpass filter of 1-30Hz. *(B)* Alpha:Beta power ratios of eyes-open and closed during 30 second trials. This setup utilized positions FZ, C3, CZ, C4, PZ, PO7, OZ, and PO8. Higher ratios were expected in the posterior region and when eyes were closed.



Figure 7. The 16-channel EEG setup during eyes-open and closed trials. (*A*) visualization of the processed EEG recording with a bandpass filter of 1-50Hz. (*B*) Alpha:Beta power ratios of all subjects with eyes-open and closed for 30 second trials. This setup utilized positions FP1, FP2, F7, F3, FZ, F4, F8, C3, C4, T5, P3, PZ, P4, T6, O1, and O2. Higher ratios were expected in the posterior region and when eyes were closed.

Α	Eyes Open vs Eyes Closed Difference				
	1 Channel	2 Channel	8 Channel	16 Channel	
Subject 1					
Subject 2			*		
Subject 3	*	*	*	*	
Subject 4			*	*	
Subject 5		*	*	*	
Subject 6			*	*	
Subject 7			*	*	

	Did Not Follow Eyes-open and Closed Difference
	Did Follow Eyes-open and Closed Difference
*	Showed Statistical Significance

B Anterior to Poster Gradient

	8 Channel	16 Channel
Subject 1	*	
Subject 2		*
Subject 3	*	
Subject 4	*	
Subject 5		*
Subject 6		*
Subject 7		*

Di Di * Sh

Did Not Follow Anterior to Posterior Gradient Did Follow Anterior to Posterior Gradient Showed Statistical Significance

Figure 8. A summary of the overall patterns seen in all channels of each setup. *(A)* The overall summary of the eyes-open and closed difference. *(B)* The overall summary of anterior to posterior gradient. The 8-channel setup showed the best pattern detection with 100% of subjects seeing at least one trend and 71% (five of seven) of them seeing both trends. The 2-channel setup showed the least trend detection with 29% (two of seven) of subjects seeing the eyes-open and closed pattern in all channels.

channel setup was the least reliable in detecting the expected patterns with only 29% showing the eyes-open and closed pattern. Paired, two-tailed T-tests were performed to determine if subjects that followed the expected pattern showed statistical significance. There were 9 instances in which the subject displayed the pattern in a certain setup but did not show statistical significance (Figure 8).

DISCUSSION

The goal of this paper was to evaluate several commonly used EEG setups and compare their potential for use in undergraduate students qualitative teaching and quantitative analysis of EEG signals. Three standard recording systems that are often used in neuroscience and physiology teaching laboratories were explored which combine affordability and ease of use. The systems selected allow for comparison of the effectiveness of multiple channels in detecting known patterns within EEG traces. This is an important consideration because cost and complexity of EEG systems increases with channel number. Medical, diagnostic, and advanced research systems may have tens or even hundreds of channels, as they are designed for precise localization of transient events. Such systems are beyond the budget of many teaching labs. The ADInstruments PowerLab 26T system has a built-in twochannel human physiological amplifier that can record EEGs. The Unicorn teaching system from g.tec has 8 channels, while the OpenBCI system contains up to 16 channels. Any of these setups were determined to be adequate in displaying the brainwaves and are relatively affordable for a teaching setting.

Overall, all setups were effective in qualitative visualization of brainwaves via counting and identifying alpha and beta waves. The 1- and 2-channel setups utilized electrode positions that allowed for a single channel view that encompassed the largest interelectrode distance. While this was efficient at simply visualizing brainwaves, quantitative analysis revealed that the setups were

unreliable in detecting the eyes closed pattern (Figure 8a). The 2-channel setup tested whether the interelectrode distance between electrodes affected the quantitative analysis of detecting the eyes closed pattern. The 1channel setup and the first channel in the 2-channel setup utilized similar electrode positions. Our results show that alpha:beta patterns to be similar in both of these trials. Furthermore, the second channel of the 2-channel setup showed some results that contradicted those of the 1channel setup and first channel. The consistency of the FPZ-OZ position in both setups, yet the differences in the FZ-PZ of the 2-channel setup indicates that interelectrode distance does affect the quantitative analysis. This is supported by the known effect of interelectrode distance on amplitude. The closer electrode positions are, the smaller the brainwave amplitude tends to be. Further distances result in larger amplitudes (Epstein et al., 1985). This effect on amplitude directly correlates with a change in power, as power scales as the square of amplitude. Due to low number and position of electrodes, the 1- and 2-channel configurations were unable to detect the anterior to posterior gradient. Therefore, they are likely to be best used for basic recording of alpha and beta waves across the cortex. These relatively simple setups allow for teaching the use of a differential amplifier, utilizing montages, selecting filters, considering best environments for the least amount of artifacts, identifying sources of electrical noise, ensuring adequate contact of electrodes to scalp, and visualizing brainwaves.

The 8- and 16-channel setups utilized higher channel numbers and different electrode positions. Therefore, both the anterior to posterior gradient and eyes opened pattern could be assessed. As far as qualitative analysis, these setups allowed for next step learning by also visualizing the anterior to posterior gradient. While the 16 channel better visualized the anterior to posterior gradient due to more electrode coverage on the head, the 8-channel setup also reliably showed this effect, and it was the most reliable in quantitative detection of the pattern (Figure 8). This could be due to the shorter leads in the 8-channel system, and the fact that the amplifier was directly attached to the head cap. This allowed for a higher signal to noise ratio in the recording. As the 16-channel setup is closer to clinical and research systems, its performance could potentially be improved by shortening the leads, and attaching the amplifier to the headcap. The 8-channel setup was the best for students to quantify the alpha and beta wave content via power spectrum analysis, but the above-mentioned modification of the 16-channel setup may allow for a better representation of the anterior to posterior gradient than in this study. Importantly, not displaying these patterns does not indicate their absence, but rather alludes to differences in the setup. Many factors within the setups can affect the EEG signal amplitude and thus the power ratio. This includes sampling rate, electrode position, number of electrodes, filters, and equipment sensitivity.

Many modules for laboratory courses incorporate hypothesis testing and statistical analysis. An EEG recording, however, is often highly variable. Brainwave amplitudes may differ several-fold across individuals, as this signal is highly dependent on head shape, skin conductivity, interference from hair, and precise placement of individual electrodes. Changes in brainwaves in response to complex stimuli are likewise individual. In research, statistical comparisons of EEGs are normally based on a large subject pool or a normative database. In a teaching lab, such practices may not be achievable. Therefore, to perform individual EEG experiments in the teaching lab that rise to the level of statistical significance in a conventional t-test or ANOVA may not be applicable. Our results reflect this by having 9 out of 30 instances in which the subjects' alpha:beta power ratios showed the expected patterns without rising to the level of statistical significance (Figure 8). Nevertheless, these statistical evaluations are useful teaching tools for undergraduate students.

This study aimed to not only determine the utility of different EEG setups for introductory and advanced lessons, but to also highlight the importance of different approaches to reading EEGs, which includes both the qualitative evaluation, as in the diagnostic field, and the quantitative approach, as utilized in research. This provides the students with a wide range of skills and real-world applications. More complex, higher density EEG recordings can certainly be useful in a teaching setting and may add capabilities, such as precise localization of event-related potentials. Our study shows the utility that can be obtained even from simpler, more easily accessible systems.

REFERENCES

- Acharya JN, Acharya VJ (2019) Overview of EEG Montages and Principles of Localization. J Clin Neurophysiol 36:325-329.
- Bazzani A, Ravaioli S, Trieste L, Faraguna Ú, Turchetti G (2020) Is EEG suitable for marketing research? A systematic review. Front Neurosci-Switz 14:594566.
- Berger H (1929) Über das Elektrenkephalogramm des Menschen. Arch Psychiat Nerven 87:527–570.
- Britton JW, Frey LC, Hopp JL, Korb P, Koubeissi MZ, Lievens WE, Pestana-Knight EM, St. Louis EK (2016) The normal EEG. In: Electroencephalography (EEG): An introductory text and atlas of normal and abnormal findings in adults, children, and infants (St. Louis EK, Frey LC eds). Chicago, IL: American Epilepsy Society.

- Epstein CM, Brickley GP (1985) Interelectrode distance and amplitude of the scalp EEG. Electroencephalogr Clin Neurophysiol 60:287–292.
- Demos JN (2019) Getting started with EEG neurofeedback, 2nd Edition. New York, NY: W.W. Norton & Company.
- Dempster J (2001) Signal analysis and measurement. In: The laboratory computer. A practice guide for physiologists and neuroscientists pp 136-171. Cambridge, MA: Academic Press.
- Feng R, Hu J, Pan L, Wu J, Lang L, Jiang S, Gu X, Guo J, Zhou L (2016) Application of 256-channel dense array electroencephalographic source imaging in presurgical workup of temporal lobe epilepsy. Clin Neurophysiol 127:108–116.
- Fetz EE (2007) Volitional control of neural activity: Implications for brain-computer interfaces. J Physiol 15:571–579.
- Ince R, Adanır SS, Sevmez F (2021) The inventor of electroencephalography (EEG): Hans Berger (1873-1941). Childs Nerv Syst 37:2723–2724.
- Kirschstein T, Köhling R (2009) What is the source of the EEG? Clin EEG Neurosci 40:146–149.
- Kopańska M, Ochojska D, Dejnowicz-Velitchkov A, Banaś-Ząbczyk A (2022) Quantitative electroencephalography (QEEG) as an innovative diagnostic tool in mental disorders. Int J Environ Res Public Health 19:2465.
- Kutluay E, Kalamangalam GP (2019) Montages for noninvasive EEG recording. J Clin Neurophysiol 36:330–336.
- Ledwidge P, Foust J, Ramsey A (2018) Recommendations for developing an EEG laboratory at a primarily undergraduate institution. J Undergrad Neurosci Educ 17:A10-19.
- Luck SJ (2022) Applied event-related potential data analysis. Davis, CA: LibreTexts, University of California.
- Miller BR, Troyer M, Busey T (2008) Virtual EEG: A software-based electroencephalogram designed for undergraduate neuroscience-related courses. J Undergrad Neurosci Educ 7:A19-25.
- Naydenov C, Yordanova A, Mancheva V (2022) Methodology for EEG and reference values of the software analysis. Open Access Maced J Med Sci 10:2351–2354.
- Noachtar S, Rémi J. (2009) The role of EEG in epilepsy: a critical review. Epilepsy Behav 15:22–33.
- Nuwer MR (2018) 10-10 electrode system for EEG recording. Clin Neurophysiol 129:1103.
- Rivera MJ, Teruel MA, Maté A, Trujillo J (2022) Diagnosis and prognosis of mental disorders by means of EEG and deep learning: a systematic mapping study. Artif Intell Rev 55:1209– 1251.
- Saha S, Mamun KA, Ahmed K, Mostafa R, Naik GR, Darvishi S, Ahsan HK Baumert M (2021) Progress in brain computer interface: Challenges and opportunities. Front Syst Neurosci 15:578875.
- Shields SM, Morse CE, Applebaugh ED, Muntz TL, Nichols DF (2016) Are electrode caps worth the investment? An evaluation of EEG methods in undergraduate neuroscience laboratory courses and research. J Undergrad Neurosci Educ 15:A29-37.
- Segawa JA (2019) Hands-on undergraduate experiences using low-cost electroencephalography (EEG) devices. J Undergrad Neurosci Educ 17:A119-124.
- Tatum WO, Olga S, Ochoa JG, Munger Clary H, Cheek J, Drislane F, Tsuchida TN (2016) American Clinical Neurophysiology Society Guideline 7: Guidelines for EEG Reporting. J Clin Neurophysiol 33:328-32.
- Tatum WO, Feyissa AM, Davis V (2018) Technical Aspects of EEG (Stern JM eds). San Diego, CA: MedLink Neurology.
- Wang Y, Nakanishi M, Zhang D (2019) EEG-Based Brain-Computer Interfaces. Adv Exp Med Biol 1101:41–65.

Received January 15, 2023, revised April 4, 2023; accepted April 4, 2023.

This work was supported by the College of Arts and Sciences, the Department of Biological Sciences and the Undergraduate Neuroscience Program at the University of Cincinnati. Furthermore, it was approved by the University of Cincinnati institutional review board as a non-human subject study (IRB #2022-0149). The authors thank the participants in the neurophysiology laboratory course.

Address correspondence to: Shelby Hatton, Undergraduate Neuroscience

Program, University of Cincinnati, 614 Rieveschl Hall, Cincinnati, OH, 45221-0006, Email: <u>Hattonsl@mail.uc.edu</u>

Copyright © 2023 Faculty for Undergraduate Neuroscience www.funjournal.org