

Assessing a learning process with functional ANOVA estimators of EEG power spectral densities

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Abstract We propose to assess the process of learning a task using electroencephalographic (EEG) measurements. In particular, we quantify changes in brain activity associated to the progression of the learning experience through the functional analysis-of-variances (FANOVA) estimators of the EEG power spectral density (PSD). Such functional estimators provide a sense of the effect of training in the EEG dynamics. For that purpose, we implemented an experiment to monitor the process of learning to type using the Colemak keyboard layout during a twelve-lessons training. Hence, our aim is to identify statistically significant changes in PSD of various EEG rhythms at different stages and difficulty levels of the learning process. Those changes are taken into account only when a probabilistic measure of the cognitive state ensures the high engagement of the volunteer to the training. Based on this, a series of statistical tests are performed in order to determine the personalized frequencies and sensors at which changes in PSD occur, then the FANOVA estimates are computed and analyzed. Our experimental results showed a significant decrease in the power of β and γ rhythms for ten volunteers during the learning process, and such decrease happens regardless of the difficulty of the lesson. These results are in agreement with previous reports of changes in PSD being associated to feature binding and memory encoding.

Keywords Neurocognitive phenomics · Electroencephalography · Brain rhythms · Power spectral density · Functional ANOVA · Learning

Introduction

A great deal of research in the area of neurosciences is being focused in developing physiological and/or behavioral measurements to provide objective and accurate quantification of the development of new abilities. Recently, a new branch of science called *neurocognitive phenomics* has been proposed by Duch (2013), which aims to explain, at least qualitatively, the phenomena that link cognitive psychology and learning with brain processes. While some of the ideas behind this new science might seem rather speculative, they serve as inspiration for our proposal.

Changes in the electroencephalogram (EEG) occurring during various mental activities in man have been the subject of many investigations since the beginning of EEG research. The use of spectral analysis for EEG has allowed determination of neurophysiological relationships (Walter 1963), as well as examination of the correlation between EEG and mental processing (Dolce and Waldeier 1974). In terms of learning, there is also much interest in analyzing the variables affecting its performance, and evidence linking spectral parameters of EEG with a number of different factors have been already reported (Harmony et al. 1990). Nevertheless, those studies so far have been more interested in the factors affecting the performance among individuals, and not on how the learning process evolves.

An early attempt to explain the correlation between EEG and learning is the one by Thompson and Obrist (1964), where the link between the active phase of learning and EEG desynchronization is first suggested. There, significant EEG changes were obtained during verbal learning as opposed to non-learning control conditions, while an over-learning condition resulted in a slight return toward control levels. However, the technical limitations of the

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time only allowed to observe such phenomena on slow brain rhythms, specifically the alpha and beta bands.

More recently, the role of faster brain rhythms in learning has been studied. In particular, the gamma band has been linked with associative (Miltner et al. 1999) and perceptual (Gruber et al. 2002) learning. In both cases, differences on a metric related to the gamma activity (coherence and induced band response, respectively) were found as probable representations of Hebbian cell assembly, which is critical for learning, memory, information processing, perception and the behavior of organisms. Still, both studies work on the basis of a learning/non-learning paradigm similar to the one by Thompson and Obrist (1964), then it is difficult to conclude about the evolution of the learning process.

Aside from that, the use of controlled elevation or suppression of specific EEG frequency components and slow potentials by means of EEG biofeedback (also referred to as *neurofeedback*) has been employed in research and clinical settings (Yucha and Montgomery 2008). The experimental application of neurofeedback protocols to healthy subjects has been shown to be a useful approach for elucidating protocol-specific effects on behavioral and electrocortical performance measures, as well as for establishing basic tenets regarding the learning process underlying the neurofeedback training rationale (Egner and Gruzelier 2001). However, while evidence delineating the effects of frequency-band neurofeedback has thus accumulated over recent years, the neurophysiological workings underlying those effects are only poorly understood. This further motivates the need of placing more effort in cultivating the area of neurocognitive phenomics.

In this paper (see also Gutiérrez et al. 2015), we propose to study the brain activity measured through EEG during the process of acquiring a new ability. Hence, we analyze the power spectral densities (PSD) of various brain rhythms as a first step to find the link between learning and brain processes. In our case, we are interested in the *evolution* of the learning process, then we analyze the influence that different difficulty levels of the task to be learned have in the PSD at different stages of the training process.

Since we are interested in the dynamics of the learning process, we require additional tools of statistical analysis that are a function of time. That is the case of functional analysis-of-variances (FANOVA) through which the effects of training on a repetition-to-repetition basis can be accounted for. Hence, the main contribution of this work is the use of the FANOVA estimators of the PSD to measure the effect of training through the process of learning, instead of the “before and after” account that the classical analysis-of-variances (ANOVA) could provide.

Experimental setup

Ten volunteers were trained through a series of twelve daily lessons to type in a computer using the Colemak keyboard layout, which is an alternative to the QWERTY and Dvorak layouts, and it is designed for efficient and ergonomic touch typing in English. Six of our volunteers were female, four male, all of them were right-handed, and their mean age was 29.3 years old with an standard deviation of 5.7 years. The lessons used during our experiment are available on-line at colemak.com/Typing_lessons. In our case, we asked the volunteers to repeat each of them five times (with resting intervals of 2 min in between). We chose Colemak touch typing as the ability to learn because most people are unaware of its existence, then it is a good candidate for a truly new ability to learn. The training process always took place in a sound-proof cubicle in which the volunteers were isolated from distractions. Hence, the volunteers were sitting in front of the computer and were engaged entirely in the typing lesson (see Fig. 1). All the experiments were carried at the same hour of the day, and all volunteers were asked to refrain of doing any additional training anywhere else. Each of the volunteers' repetitions were chronometred in order to assess the improvement in their ability to type. Note that this included the time used to correctly write all the words in the lesson, as well as the time consumed in erasing and rewriting words due to any mistakes.

Each of the volunteers were subjected to four sessions of EEG recordings, all of which were acquired using the B-Alert X10 wireless system from Advanced Brain Monitoring, Inc., which includes a combination of nine EEG sensors with fixed gain referenced to linked mastoids and located at mid-line and lateral positions. Those sensors, based on the international 10–20 reference system, are positioned at F3, Fz, F4, C3, Cz, C4, P3, POz, and P4. All data are acquired at a sampling frequency of 256 Hz and filtered with a fifth-order Butterworth bandpass filter with

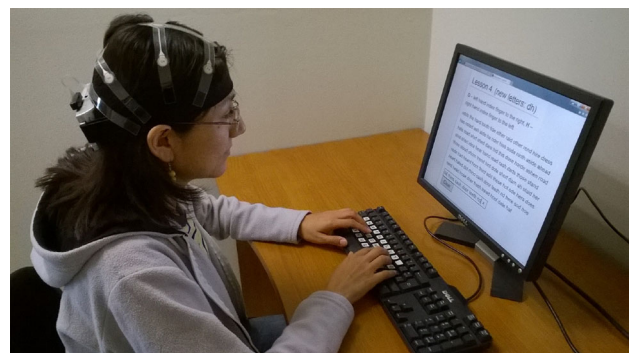


Fig. 1 Experimental setup

cutoff frequencies at 0.1 and 100 Hz. Our measurements were acquired as follows:

- An initial EEG measurement is performed before starting the training sessions in order to define a baseline state through B-Alert's *Alertness & Memory Profiler* (AMP), which corresponds to a comprehensive neurocognitive test battery that synchronizes B-Alert EEG data to evaluate and compare metrics across populations and interventions (Berka et al. 2010).
- EEG measurements were acquired during the repetitions of the typing task at the fourth, eighth, and eleventh lessons, in which one-, two-, and three-full horizontal lines of keys in the keyboard were used, respectively. Hence, those three lessons are assumed as representative of low-, medium-, and high-level difficulty in performing the task. During the experiment, the volunteer typed while seated in front of the computer and the corresponding lesson was displayed in the monitor.

The B-Alert software provides a second-to-second estimate of the EEG's mean PSD for the brain rhythms in the following ranges: 1–2 Hz for δ , 3–7 Hz for θ , 8–12 Hz for α , 13–29 Hz for β , and 30–40 Hz for γ . The PSD is computed by performing fast Fourier transform (FFT) on a segment of data that is of interest, and calculating the amplitudes of the sinusoidal components for the frequency bins defined previously. The second-to-second estimates of the PSD are the result of applying a 50 % overlapping window which averages the PSD across three overlays to smooth the data. Each overlay contains 256 data points with 128 being shared for each overlay. A Kaiser window is applied to each overlay in order to accentuate the contribution of power from the signal in the middle third of the overlay and minimize the impact of signal near the edge of the overlay. Prior to computing the PSD, the raw signals are processed to eliminate spikes, excursions and amplifier saturation, as well as excessive electromyographic (EMG) contribution. In particular, a combination of high frequency (based off 70–128 Hz for each overlay) and low frequency (based off 35–40 Hz) detectors are used to identify periods with excessive EMG. If only one overlay has EMG, then the PSD for the current epoch is based on the average of the remaining two overlays. If excessive EMG is found in two overlays, the epoch is excluded from the analysis.

Furthermore, the B-Alert software also provides a probabilistic measure of the cognitive state based on the results of the AMP. Hence, in addition to the PSD values, each one-second epoch is assigned one of the following states: *sleep onset*, *distraction*, *low engagement*, and *high engagement*. More information about the validity of this metric can be found in the paper by Johnson et al. (2011).

Proposed analysis

We propose to perform different statistical tests in order to establish a relationship between changes in PSD during the learning task and (1) frequency of the brain rhythm, (2) EEG sensors, and (3) repetitions of a lesson. Hence, our proposed analysis consists in two-sample t tests, two-way ANOVA, and the calculation of the FANOVA estimators, respectively.

Two-sample t tests

We perform two-sample t tests to check if the PSD estimates from the measurements at the last repetition are significantly different in comparison to those from the first. The statistical tests are performed for all nine measuring channels at the three measured lessons, but only the PSD epochs where the B-Alert software classified the volunteer's cognitive state as *high engagement* are considered. Under those conditions, the test statistic under the null hypothesis has a student's t distribution with $N_1 + N_5 - 2$ degrees of freedom, where N_1 and N_5 are the number of epochs classified as *high engagement* during the first and last repetition, respectively. The t tests are performed for each subject and for a specific brain rhythm in order to determine the frequency band in which changes in PSD are linked to the learning task. Only those frequencies for which a majority of sensors show significant change in PSD would be considered for the next statistical test through ANOVA.

Two-way ANOVA

In order to get a better understanding of the relationship between changes in PSD as result of the learning task, we propose to find if the $I = 5$ repetitions and the $D = 3$ difficulty levels of the tasks have a mixed interaction with changes in the PSD. Hence, for each subject, we perform nine (one for the data acquired at each sensor) two-way ANOVA tests. Those sensors in which a significant mixed effect of the factors (repetitions and difficulty) is found will be selected as a personalized set to be used in the following functional test.

Based on those conditions, the test statistic under the null hypothesis has a F distribution with $(I - 1)(D - 1)$, $ID(N_{i,j} - 1)$ degrees of freedom, where $N_{i,j}$ corresponds to the number of epochs classified as *high engagement* in the i th repetition and j th difficulty level. Given that $N_{i,j}$ is not a constant value, the two-way ANOVA tests here proposed are unbalanced (Fujikoshi 1993). At the end, the personalized sets will be formed with those $K = 6$ sensors for which the probability of the test statistic is much more smaller than the significance level assumed (for details behind this reasoning, see Salazar-Varas and Gutiérrez 2015).

FANOVA estimators

With the selected frequencies and sensors in which significant changes in PSD are observed, the final step in the analysis is to assess the evolution of those changes as a function of the repetitions. In this way, we will be able to get some insight about the evolution of the brain rhythms as the process of learning happens. In order to achieve that, we propose to analyze the FANOVA estimators obtained from the data at each lesson.

FANOVA is a “functionalized” version of the standard ANOVA test, in which the observations $y_{i,k}$, for $i = 1, 2, \dots, I$ repetitions, and $k = 1, 2, \dots, K$ sensors, are modeled by a fixed-effect ANOVA model (Ramsay 2006):

$$y_{i,k}(n) = a_0(n) + a_i(n) + \varepsilon_{i,k}(n), \tag{1}$$

for $n = 1, 2, \dots, N$, where $a_0(n)$ is the *main-effect* function, $a_i(n)$ are the *treatment* functions, and $\varepsilon_{i,k}$ are independent zero-mean Gaussian errors. In order for the treatment functions to be uniquely identifiable, they have to satisfy the constraint $\sum_i a_i(n) = 0$ for all n .

In our case, the idea behind using (1) is to represent the PSD as having a common component, as well as an element of extra variation due to the effect of the i th repetition. Under those conditions, the functions $a_0(n)$ and $a_i(n)$ are estimated as follows:

$$\hat{a}_0(n) = \frac{1}{IK} \sum_{i,k} y_{i,k}(n), \tag{2}$$

i.e., the average PSD across all repetitions and sensors, and

$$\hat{a}_i(n) = \bar{y}_i(n) - \hat{a}_0(n), \tag{3}$$

with $\bar{y}_i(n) = \frac{1}{N_i} \sum_k y_{i,k}(n)$. The FANOVA estimators in (2) and (3) are those that minimize the residual sum of squares:

$$\begin{aligned} \min_{a_0, a_i} \sum_n \sum_{i,k} [y_{i,k}(n) - (a_0(n) + a_i(n))]^2, \\ \text{subject to } \sum_i n_i a_i(n) = 0. \end{aligned} \tag{4}$$

The problem in (4) corresponds to minimize the discrete version of the least-mean-squared sum of errors (LMSSE). More details on the solution and some computational caveats involved in (4) are discussed in Ramsay’s (2006) book.

Results

Before proceeding with the statistical tests proposed in “Proposed analysis” section, we looked at the time it took our volunteers to complete each repetition. We denote this time at the i th repetition as t_i , for $i = 1, \dots, 5$. The mean \pm standard deviation for the first repetition in the fourth,

eighth, and eleventh lessons were 454.2 ± 103.62 , 461.9 ± 114.22 , and 465.5 ± 129.01 s, respectively. By the fifth repetition, those times got reduced to 306.8 ± 48.20 , 348.9 ± 77.62 , and 380.5 ± 120.54 s, respectively. Hence, as a measure of the percentage improvement, we compared t_1 and t_5 for all lessons through the following formula:

$$\text{Improvement} = 100 \left(1 - \frac{t_5}{t_1} \right). \tag{5}$$

Therefore, a faster last repetition in comparison to the first one is considered as an improvement. In our case, such improvement is assumed to be a reflection of the volunteer’s learning process, and the results can be seen in Fig. 2. There, we note that the improvement gets reduced as the difficulty in the lessons increase. However, there was always an improvement on average for our subjects at all lessons.

Next, the t tests proposed in “Two-sample t tests” section were applied to the data of each sensor in order to check if the PSD at the first and last repetition of a lesson were significantly different. Note that the percentage of PSD epochs classified as *high engagement* was always between 52 and 72 % and, for each volunteer, the standard deviation of that value was never above 5 % between repetitions of the same lesson, then we can guarantee the commitment of the subjects to the experiment and discard tiredness effects. An example of the data used for this analysis is shown in Fig. 3. A total of $(10 \text{ subjects}) \times (9 \text{ sensors}) \times (3 \text{ levels of difficulty}) = 270$ two-sample t tests were performed for each of the brain rhythms of interest. In all cases, the tests were implemented with the *Statistics and Machine Learning Toolbox*TM of Matlab (www.mathworks.com), and a significance level of 0.01 was used. As result, the number of those tests (in percentage) that rejected the null hypothesis

$$H_0: \overline{\text{PSD}}_1 = \overline{\text{PSD}}_5, \tag{6}$$

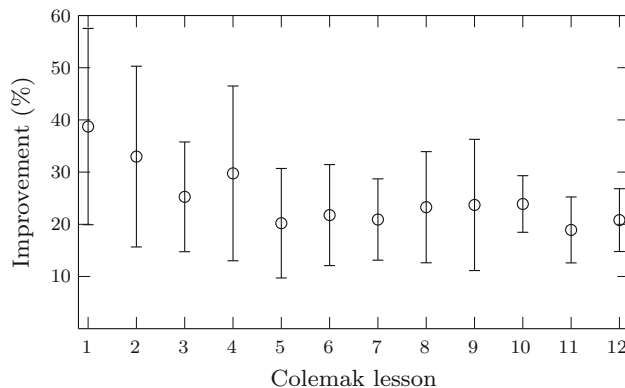


Fig. 2 Average improvement in the lesson’s execution time for our ten subjects. Error bars indicate the mean \pm standard deviation

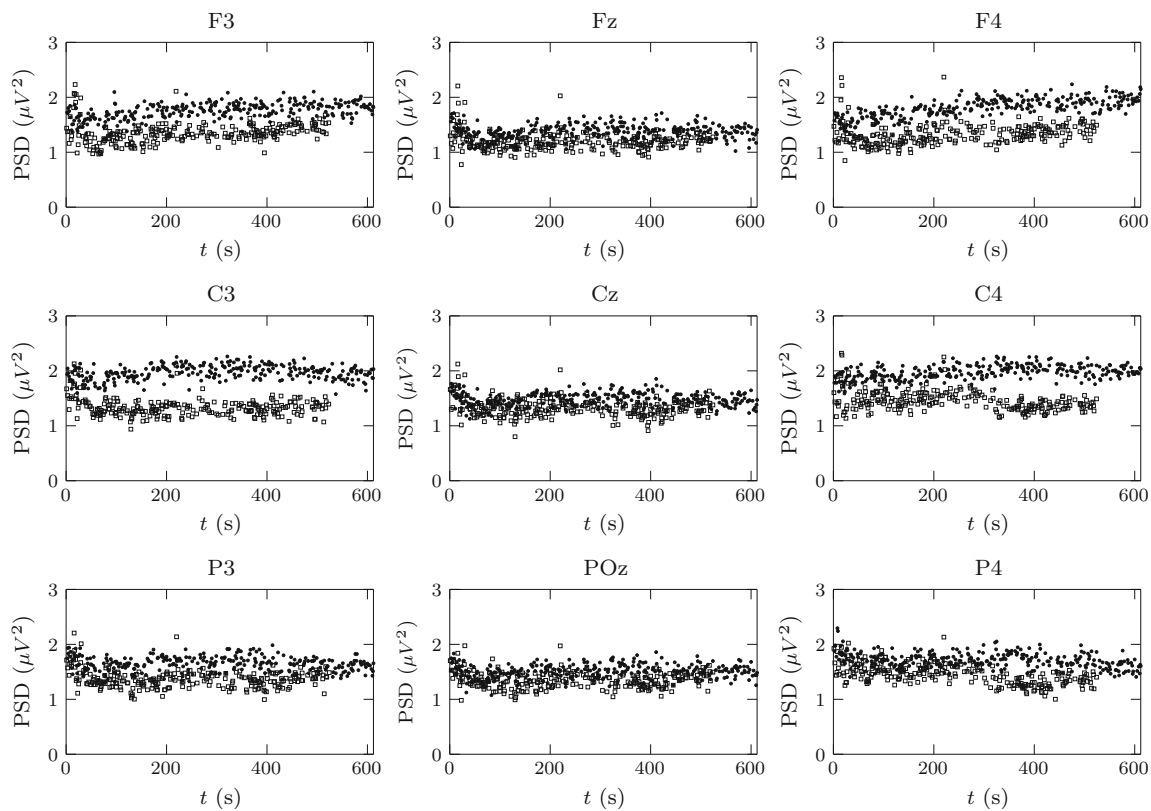


Fig. 3 PSD estimates of the γ rhythm while Subject 5 performed the eleventh Colemak lesson. Points marked with circle correspond to the first attempt of that lesson, while those indicated as square correspond to the fifth

where $\overline{\text{PSD}}_{\{*\}}$ indicates the mean PSD at a given repetition, turned out to be: 18.89 % for δ , 27.41 % for θ , 31.11 % for α , 54.44 % for β , and 71.11 % for γ . Based on those results, our following analysis focused in the β and γ bands, for which the change was such that $\overline{\text{PSD}}_5 < \overline{\text{PSD}}_1$ in most of the t tests (for more details, see Gutiérrez et al. 2015).

Two-way ANOVA tests were performed on the PSD of the selected brain rhythms. The aim of these tests was to select those EEG sensors in which interaction between repetitions and difficulty level was observed. Again, the tests were implemented with Matlab, and a significance level of 0.01 was used. Overall, $(9 \text{ sensors}) \times (2 \text{ frequency bands}) = 18$ two-way ANOVA tests were evaluated for each subject’s data. As result, $p \ll 0.01$ in most tests, then the $K = 6$ sensors with the lowest p -values were selected for the following analysis. However, some cases were above the significance level and the corresponding sensors had to be excluded from the following tests (we even had a couple of cases where $K < 6$). A list of such cases is shown next, in which the pair of $[F, p]$ values of the corresponding two-way ANOVA test are included as well:

- Cz [1.60, 0.119], C4 [1.94, 0.050], POz [2.28, 0.019], and P4 [2.10, 0.032] in the β band were excluded for Subject 1;

- Fz [1.94, 0.051] and F4 [4.06, 0.043] in the β band were excluded for Subject 6;
- C3 [1.55, 0.137] and Cz [2.36, 0.016] in the β band, and C4 [2.08, 0.034], P3 [1.03, 0.412], and P4 [2.39, 0.015] in the γ band were excluded for Subject 8;
- F3 [1.35, 0.215], Fz [1.42, 0.183], C3 [2.29, 0.019], Cz [2.09, 0.034], P3 [2.38, 0.015], and POz [2.48, 0.011] in the β band were excluded for Subject 9.

It is worth mentioning that Subject 8, who was the only one that had sensors excluded in both bands, had experience in formal typing using the QWERTY layout.

Once the personalized sets of sensors had been chosen, we proceeded to compute the FANOVA estimators as described in “FANOVA estimators” section, but with two additional processing steps that were applied to the PSD:

1. In (1), it is assumed that the samples are equally spaced, but that is not our case due to the constraint of only using PSD values that corresponded to a cognitive state of *high engagement*. Therefore, we applied a spline interpolation to the PSD in order to have uniformly spaced samples.
2. In our case, $N_1 \neq N_2 \neq \dots \neq N_5$, then an unbalanced FANOVA test would have been required. Instead of that, we decided to “balance” the data by obtaining

new time series corresponding to autoregressive (AR) processes fitted from the original PSD values through the method proposed by Schneider and Neumaier (2001). Hence, our data were both fitted and re-synthesized with third-order AR models and the new time series had a number of samples $N = \max\{N_1, N_2, \dots, N_5\}$.

Under those conditions, we obtained the FANOVA estimators for each subject, at each difficulty level, and for the

selected frequency bands. An example of the result of this process is shown in Fig. 4. There, we can notice that the main difference between treatment functions is on the mean, which is computed as $\bar{a}_i = \frac{1}{N} \sum_{n=1}^N \hat{a}_i(n)$.

A comparison of all the values of \bar{a}_i for our group of subjects is shown in Fig. 5. Note that each of the values there actually correspond to the grand average of \bar{a}_i over 100 independent realizations of the synthesized data in order to minimize the effects of the variability introduced by the AR

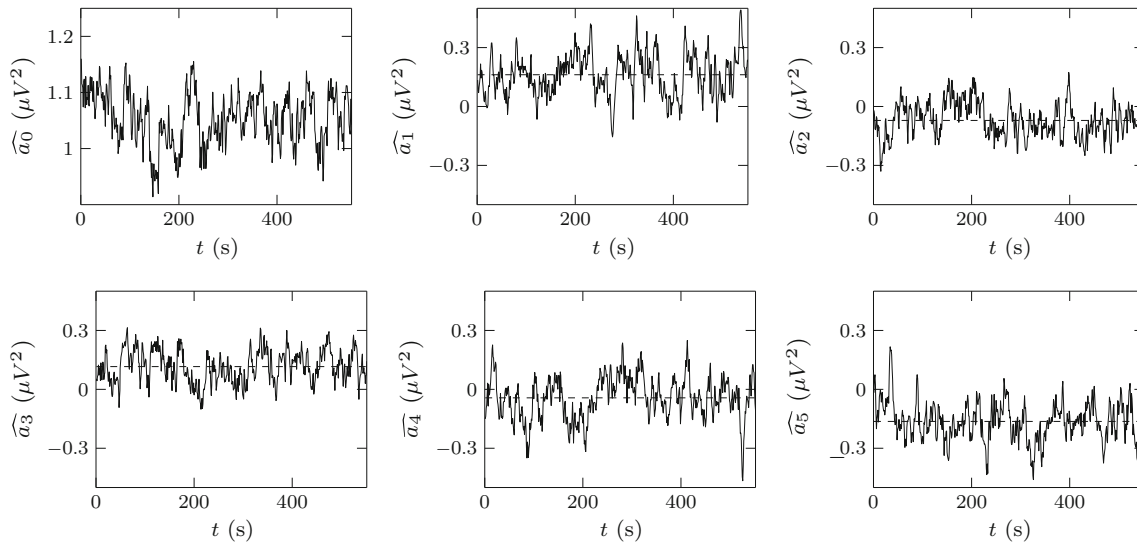


Fig. 4 Example of FANOVA estimators for one subject at a given difficulty level. The dashed lines represent the mean value \bar{a}_i of the corresponding treatment function

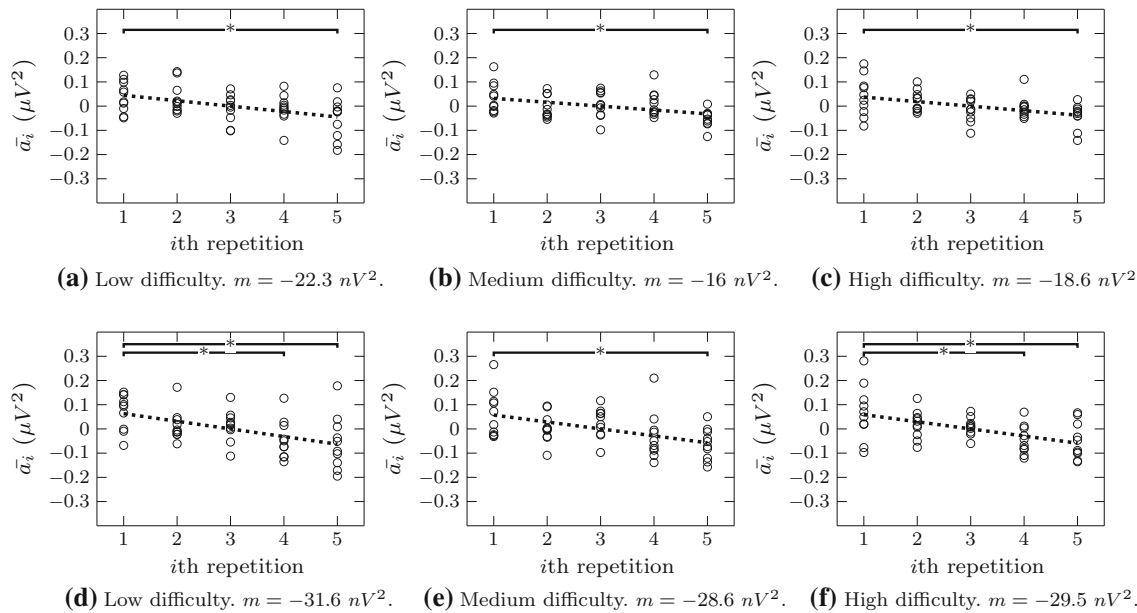


Fig. 5 Mean effect of training for our ten subjects according to the proposed analysis of the FANOVA estimators. The top row (a–c) corresponds to β band, while the lower row (d–f) corresponds to γ band. The asterisks indicate those repetitions at which the mean PSD

of the subjects is significantly different than the one obtained at the first repetition ($p < 0.05$). The dotted lines correspond to the linear regressions of the data, for which the corresponding slope (m) is indicated below each graph

modeling process in our results. Furthermore, we performed one-way ANOVA and multiple-comparison tests in order to assess the effect in our subjects of repeating the lesson. As result, we found a significant decrease ($p < 0.05$) in the β and γ rhythms from the beginning to the end of a lesson, and this result was independent of the level of difficulty. The speed of such decay in power was approximated by the slope (m) of the linear regression of the data.

Finally, in order to highlight the fact that the decrease in the power of β and γ rhythms is indeed related to an improvement in the ability to type, we observed the relationship between the values of \bar{a}_i and the time t_i required by all subjects to complete the tasks for the case of the first and last repetition (i.e., $i = 1, 5$), as those were the repetitions for which all the difficulty levels turned out with significant decrease in β and γ , and those were the times we considered for our measure of improvement shown in Fig. 2. Therefore, we computed the Pearson's correlation of the pairs (\bar{a}_i, t_i) , and the results shown in Fig. 6 indicate that there exists a moderately strong to strong correlation between FANOVA estimators and time required in the first and last repetitions, such that positive values of \bar{a}_i correspond to larger times to execute the task, while negative values correspond to shorter times.

Discussion

The main purpose of introducing a functional analysis of the PSD was motivated by the desire of having a tool capable of describing the time-changing dynamics of the

learning process, then the FANOVA estimators were proposed for such task. Perhaps our case of study was not the best example to show the full potential of this type of analysis, given that the observed changes turned out to be only on the mean of the PSD. Nevertheless, we believe that our analysis can be seen as a proof-of-the-concept on how to proceed in the case of more elaborated learning tasks and with longer training periods in which non-stationarity of the processes to be analyzed could be expected.

Now, in terms of the decrease we found in the β and γ rhythms, we strongly believe they can be the result of two phenomena previously observed in the context of (1) motor learning and reduced preparatory or attentional demands, and (2) memory encoding, respectively.

In the first phenomenon, Niemann et al. (1991) reported a decrease of the EEG negativity which was correlated with the increase of motor skill (in their case, of a complex finger motor task). More recently, Erbil and Urgan (2007) showed the correlation between the decrease in β amplitude as result of long-duration repetitive hand movements similar to those studied by Niemann et al. (1991). It might seem, therefore, possible to explain the β suppression observed in our typing task as the result of the sustained motor activity having the habituation effect of the cortical mechanisms stimulated by the somatosensory stimuli related to the learning task.

In addition to the β decrease, multiple studies in humans suggest a role of induced γ oscillations in coupling perception and learning. That is the case of memory encoding that, in its simplest form, has been shown to appear as a

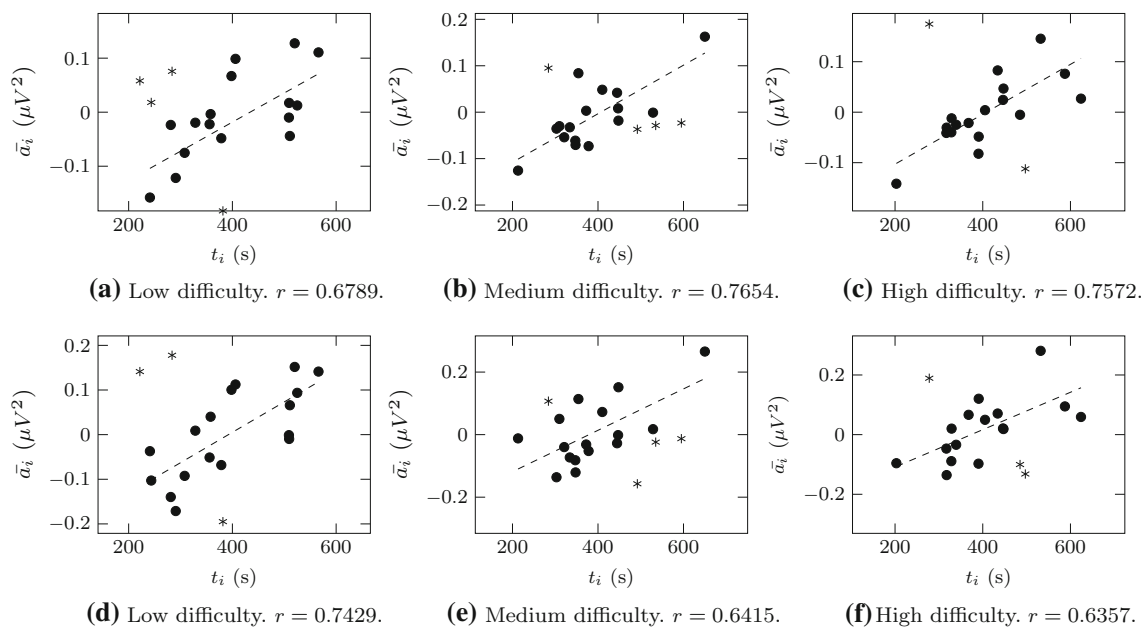


Fig. 6 Values of (\bar{a}_i, t_i) for all subjects and for $i = 1, 5$. The *top row* (a–c) corresponds to β band, while the *lower row* (d–f) corresponds to γ band. The Pearson's correlation coefficient r is given for each band

and difficulty level (with $p < 0.01$ for all cases). Data point considered as outliers are indicated by *asterisk*

decrease of neural activity upon repeated presentations of the same stimulus. This phenomenon, known as *repetition suppression*, can be specifically observed in the γ rhythm (Gruber and Miller 2005). Upon repeated presentation of a stimulus, the synaptic plasticity fostered by γ oscillations would help selecting the most relevant processes and lead to the creation of a sparse and faster neural route, resulting in a sharpened representation of the learned stimulus. Although repetition suppression might explain the reduction in γ we observed, further analysis is necessary, as different components of the γ rhythm (in frequency and space) have been related to either the perceptual or the overall attentional allocation of the stimuli in memory (Chaumon et al. 2009).

Conclusions

We proposed an analysis framework based on multiple statistical tests for EEG data acquired during the process of learning a task, in this case typing in Colemak. However, it is through the use of the FANOVA estimators of the PSD that we propose to get an insight on the effects of different factors (amount of training and difficulty) that the process of learning has in the brain dynamics. We showed the feasibility of examining sustained EEG activity and reinforced the hypothesis that some of its rhythms correlate to the process of learning. Our results were in agreement with previous evidence of the involvement of β and γ rhythms in the process of acquiring a new ability.

In this paper we focused our attention in the decreasing trend of the β and γ rhythms given that such trend was more obvious in those bands through our preliminary analysis (Gutiérrez et al. 2015). However, a less evident increase in the α band could be observed in some subjects and in some sensors. Then, we believe it is important also to study this band and its possible interaction with β and γ as others have previously suggested (Canolty and Knight 2010). We strongly believe that the analysis framework here presented has the potential to help to understand the nature of such coupling at different stages of the learning process. Furthermore, the balance between increasing α and decreasing β and γ could be studied as the learning task progresses through a combination of the methods here proposed and time-varying AR analysis as the one proposed by Gutiérrez (2013).

Finally, it is also important to study the spatial distribution of the different rhythm's power in order to better understand the underlying brain processes (Miltner et al. 1999). Therefore, for our future work, we plan to acquire a greater number of EEG sensors, as well as to introduce an analysis of EEG coherence similar to the one proposed by

Salazar-Varas and Gutiérrez (2015), but for the case of studying a learning task.

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