EEG band power analysis corresponding to salivary amylase activity during stressful computer gameplay

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Abstract

The cortisol and norepinephrine from human salivary can represent psychological conditions. A portable salivary amylase monitor device (sAA) has existed; however, how the sAA corresponds to the central nervous system changes is still limited to carry out. Twenty university students aged between 20 and 22 years participated in which they played a stressful computer game during the experiment. Nineteen EEG electrodes were attached to the head scalp while the relative power on the delta, theta, alpha, and beta-band was calculated. The sAA value was obtained using a portable device called Nipro Cocorometer from Japan. The sAA levels and the brain's relative band power increased. Beta waves of the brain's right hemisphere were found higher than that of the left hemisphere, especially on the right temporal (T4, p < 0.01). Then, we concluded that the beta-band power on the right hemisphere corresponds to the sAA changes.

Keywords: stress; salivary amylase; EEG; beta-band power; cocorometer

1. Introduction

Currently, mental stress caused comes from various sources, including information, social media, and technology [1,2]. Stress can be defined both as a reaction of a human to external stimuli (stressors) as a non-specific response of the body to any noxious stimulus [3,4]. It can be recognized by the rising cortisol and norepinephrine in the blood [5,6]. From this, stress can be classified into human body stress and cell stress, psychological and physical stress, and eustress or distress. Eustress is commonly known as a positive psychological state; while distress is known as a negative one [7].

Currently, many studies report how bio-signals from sensors represent mental health and other psychological conditions [8]. Most applied technologies use heart rate and blood pressure to estimate the psychological state [9-11]. However, the heart rate and blood pressure are markedly affected by homeostasis, and those indices are ambiguous during a direct comparison to ordinary and other specific conditions [12]. On the other side, quantifying the hormones such as cortisol and norepinephrine is able to distinguish between eustress and distress. It has been utilized to validate mental health studies objectively. Despite the ability to differentiate different mental states, cortisol and norepinephrine are problematic to be readily quantified and uneasy to observe within a short period [5,13,14].

To overcome the time limitation of quantifying the hormones, Yamaguchi et al. established a method to observe the salivary alpha-amylase activity also known as salivary amylase activity (sAA) through a portable device to substitute the cortisol assessment. Here, the results can be obtained by using the sAA to quantify the stresor levels only after one or a few minutes of data retrieval rather than conventional methods [12,15]. Previous studies that quantified sAA using a portable device to assess the mental state are already widely used such as to evaluate the mental stress of breast cancer patients, the effect of thermal comfort, evaluate the pain measurements, and treat depression [8,16].

Since heart rate and blood pressure have few limitations to differentiate the diversity of mental health, such as eustress and distress, it is required to find out bio-signals that follow the sAA’s changes well. It is already well-known that the brain is the central organ controlling the whole system of the human body. According to the stress itself, the brain is responsible for perceiving and adapting to social and physical stressors caused by multiple interacting paths from the cell to the surface to the cytoskeleton to epigenetic regulation and non-genomic mechanisms [17]. According to previous research, the prefrontal cortex is crucial in signaling the stress pathway [18]. EEG is commonly utilized to measure changes in neurophysiological activity linked with specific stimuli or assignments in large-scale studies of stress detection utilizing biosignals [19]. According to the study, the prefrontal cortex contributes to discriminate between distinct mental states. Nonetheless, the consensus identified conflicting band power patterns (increase or decrease) employed in stress detection
studies and the way they significantly alter during an unpleasant or stressful state. Compared to the beta band, the majority stated that a decrease in alpha is the greatest way to discern between two mental states. To induce stress conditions, most of stress detection investigations have employed fundamental treatments such as mental arithmetic tests, Stroop tests, visual and audio stimuli, IAPS photographs, driving tests, and social stress tests. Therefore, it is believed that the use of such unusual stimuli can strengthen the study of stress detection and contribute to the addition of evidence for the study of mental states [19,21].

Regarding the review of studies on the detection of stress, it can be found out that stress while computer gaming remains limited. According to a prior study, the music in a video game is the cause of tension when playing the game. Based on cortisol level validation, it has been found that the temporal region contributes to differentiate the stress condition [22]. One of the most recent and relevant studies was conducted utilizing EEG to identify changes in mental state while playing easy, medium, and difficult games [23]. Since accuracy can still be enhanced, our proposed research can contribute to a deeper comprehension of stress detection. Moreover, evaluating stress levels using sAA is less expensive and simpler than measuring stress appearance with an ELISA kit based on cortisol levels [15]. Determining if brain wave activity can predict stress circumstances following the sAA level necessitates the expansion of past research. As a result, our suggested study to determine how the reflection of sAA corresponding to the central nervous system can provide valuable information.

Electroencephalograph (EEG) signals reflect the electric activities of the brain. It has a major role in most studies on the brain in various fields such as medicine, pathological conditions, and cognitive markers, including the science of the game [24]. The ability to provide a better temporal resolution of brain activity successfully improves the study of psychological stress using EEG. This study aims to examine the following aspects: (1) to find out whether the brain waves from the frequency decomposition could reflect the biochemical process of sAA changes during different phases (resting to intense activity/playing a computer game); (2) to confirm this study with previous findings and the way it can improve the understanding of the human's body properties using bio-signals, especially to surrogate the validity and utilization of brain waves instead sAA to assess mental health; and finally (3) to extend the study of EEG to quantify the hormonal changes and/or biochemical processes that represent a psychological condition using a specific index.

2. Materials and Methods

2.1. Ethical consideration

This study was conducted in accordance to the principles of the Declaration of Helsinki. Also, this study has already been approved by the Committee of Ethics of Faculty of Medicine, Universitas Islam Indonesia Yogyakarta. All data already obtained the informed consent of the subjects and were kept confidential and anonymous.

2.2. Subjects

This study enrolled twenty university students aged between 20 and 22 with an average Body Mass Index (BMI). Due to gender differences in brain activity recording, we avoided cross-gender participants (males × females) [25]. Prior to the experiment, each subject should obtain informed consent. Subjects were advised to acquire adequate sleep, maintain a healthy weight, and abstain from any medication treatment. Also, this experiment must verify that the participants never performed a similar task or assignment to retain the research's objectivity. To ensure an objective evaluation, we ensured that the participants were unfamiliar with the game prior to the experiment. If it was discovered that they had already conducted it, the subject was discarded.

2.3. Experimental design

Figure 1 shows the experimental setup and procedure to stimulate psychological changes such as mental stress. Each subject must sit and relax in front of a 22-inch computer display. The range between the display monitor and the subject's eyes was 40-60 cm. The participants were asked to rest before the experiment started (five minutes). Then, we asked them to play a game called "Syobon Action," which had an extreme difficulty for ten minutes and was frustrating [26]. The player took control of a kitten-like character (resembling Toro Inoue) who must navigate through side-scrolling platform levels reminiscent of Super Mario Bros. The game was divided into four levels (six in the online version), each of which had traps designed to deceive the player and exploit their prior

![Fig. 1. The experimental setup and procedure](image-url)
knowledge of Super Mario Bros. The gameplay difficulties are the most frustrating and stressful aspect of the experiment. The game gets stressful because the gameplay challenges the player to restart and try the game unlimited to pass all the obstacles without any limitation. The preview of the game is shown in figure 2.

The player must play the game three times for a total of thirty minutes. Each subject must then sit quietly for five minutes before the experiment was seen complete. In general, the experiment consisted of five distinct phases. 1) Pre-experiment (5 mins); 2) Game 1 (10 mins); 3) Game 2 (10 mins); 4) Game 3 (10 mins); and 5) post-experiment (5 mins). Between the segment junctions, we observed the salivary amylase activity (sAA). While, we captured brain activity between the first and fifth segment.

2.4. Data recordings

We used a portable salivary amylase monitor device from Cocorometer Nipro Japan to collect the saliva from participants. As depicted in figure 1, we collected it five times. We recorded the brain activity from the subject's head scalp using 10-20 nineteen channels EEG from Mitsar EEG, sampled the EEG data to 250 Hz and maintained the impedance under 10 KOhm. The data were recorded by a software called a EEG Studio where the sample EEG data generated from each band power. For analysis purposes, we divided the relative band powers corresponding to the experiment's segments. The primary legacy of this study stated that relative band powers provided more accurate results to neurologic disease detection than absolute band powers [31]. Another issue was also due to physiological differences during EEG recording that differed the absolute powers of each person during the same recording environments. A study of stress detection using bio-signals also agreed that EEG's relative power is widely used for extracting intra-individual changes corresponding to the stress response [19]. Thus, relative power could be the most objective parameter to quantify brain properties to indicate mental state changes.

2.6. Data analysis

Since we tended to indicate the uncertainty around the estimate of the mean measurement rather than how widely scattered some measurements are, we presented the data of this study by using mean and standard error (SE). Since the data were not distributed normally, we established a non-parametric statistical method called as Mann Whitney U test to discriminate the brain and salivary amylase activity between the experiment's junctures. We used a test value (p-value) less than 0.05. In other
We considered the data is significantly different while the p-value was less than 0.05. We also employed the beta analysis regarding the category of AA vs. low AA levels (≤ 30 kU/l). The data analysis was done using Python and R programming languages.

### Table 1. The sAA and relative band power pre- and post-experiment (*p<0.05, **p<0.01)

| Parameter | BEFORE | AFTER | BEFORE | AFTER | BEFORE | AFTER | BEFORE | AFTER | BEFORE | AFTER |
|-----------|--------|-------|--------|-------|--------|-------|--------|-------|--------|-------|
| Alpha-Amylase Saliva (kU/L) * | 8.5000 | 1.8855 | 13.8000 | 2.6194 |
| Relative Band Power | Delta | Theta | Alpha | Beta |
| | Mean | SE | Mean | SE | Mean | SE | Mean | SE | Mean | SE |
| 1 Fp1-Av** | 0.4915 | 0.0209 | 0.4791 | 0.0174 | 0.1633 | 0.0056 | 0.1736 | 0.0053 | 0.1074 | 0.0079 |
| 2 Fp2-Av* | 0.4740 | 0.0208 | 0.4655 | 0.0185 | 0.1598 | 0.0057 | 0.1685 | 0.0058 | 0.1099 | 0.0081 |
| 3 F7-Av | 0.4059 | 0.0187 | 0.3939 | 0.0167 | 0.1489 | 0.0061 | 0.1497 | 0.0059 | 0.1248 | 0.0082 |
| 4 F3-Av | 0.3132 | 0.0168 | 0.2997 | 0.0130 | 0.1432 | 0.0069 | 0.1413 | 0.0056 | 0.1523 | 0.0090 |
| 5 F3-Av* | 0.2913 | 0.0120 | 0.2845 | 0.0110 | 0.1570 | 0.0069 | 0.1568 | 0.0051 | 0.1784 | 0.0091 |
| 6 T4-Av | 0.2945 | 0.0139 | 0.2971 | 0.0153 | 0.1423 | 0.0057 | 0.1407 | 0.0051 | 0.1562 | 0.0079 |
| 7 F8-Av | 0.3766 | 0.0144 | 0.3801 | 0.0164 | 0.1486 | 0.0048 | 0.1468 | 0.0045 | 0.1281 | 0.0064 |
| 8 T3-Av | 0.2991 | 0.0187 | 0.3123 | 0.0166 | 0.1311 | 0.0073 | 0.1363 | 0.0067 | 0.1308 | 0.0078 |
| 9 C3-Av | 0.2916 | 0.0204 | 0.2782 | 0.0161 | 0.1382 | 0.0059 | 0.1377 | 0.0061 | 0.1692 | 0.0113 |
| 10 Cz-Av | 0.3204 | 0.0179 | 0.3189 | 0.0155 | 0.1572 | 0.0050 | 0.1604 | 0.0047 | 0.1730 | 0.0117 |
| 11 C4-Av | 0.3062 | 0.0227 | 0.2994 | 0.0191 | 0.1382 | 0.0057 | 0.1418 | 0.0061 | 0.1728 | 0.0135 |
| 12 T4-Av | 0.3042 | 0.0227 | 0.3034 | 0.0204 | 0.1274 | 0.0066 | 0.1315 | 0.0078 | 0.1220 | 0.0068 |
| 13 T5-Av | 0.3338 | 0.0194 | 0.3308 | 0.0170 | 0.1457 | 0.0061 | 0.1497 | 0.0058 | 0.1574 | 0.0101 |
| 14 P3-Av | 0.3288 | 0.0219 | 0.3273 | 0.0182 | 0.1394 | 0.0057 | 0.1420 | 0.0058 | 0.1722 | 0.0131 |
| 15 Pz-Av | 0.3437 | 0.0201 | 0.3458 | 0.0178 | 0.1473 | 0.0063 | 0.1493 | 0.0059 | 0.1763 | 0.0132 |
| 16 P4-Av | 0.3350 | 0.0219 | 0.3322 | 0.0189 | 0.1405 | 0.0066 | 0.1436 | 0.0065 | 0.1745 | 0.0140 |
| 17 T6-Av | 0.3341 | 0.0203 | 0.3322 | 0.0174 | 0.1425 | 0.0060 | 0.1457 | 0.0058 | 0.1614 | 0.0117 |
| 18 O1-Av* | 0.3239 | 0.0204 | 0.3228 | 0.0174 | 0.1373 | 0.0067 | 0.1436 | 0.0068 | 0.1524 | 0.0116 |
| 19 O2-Av | 0.3394 | 0.0188 | 0.3260 | 0.0188 | 0.1417 | 0.0061 | 0.1429 | 0.0063 | 0.1512 | 0.0110 |
participants to find out whether the stressful gameplay was able to increase their salivary levels. The higher levels of salivary amylase represented the higher mental stress. Here, we found that the sAA value was higher after playing a game (13.80 ± 2.62 kU/l) compared to before (8.50 ± 1.88 kU/l) without any significant difference (p > 0.05).

Brain activity exhibited the same pattern in lower (theta; 4-7 Hz) and higher frequency bands (beta band; 13-30 Hz). Table 1 shows that in prefrontal cortex, the Fp1 and Fp2 sites demonstrated the increasing theta band (4–7 Hz) relative power from 0.1633 to 0.1736 (p = 0.0075) and 1.1598 to 1.1685 (p = 0.0191), respectively. On the other side, the occipital lobe (O1) revealed a substantial increase in theta power from 0.1373 to 0.1436 (p = 0.0352). Furthermore, the beta band (13 – 30 Hz) showed the same rising trend relative power on the Fz site with the elevations of 0.0105 before and after the experiment (p < 0.05).

Regarding the pre-and post-experiment, we found that brain activity mostly showed no difference before and after playing the game corresponding to the sAA activity. Therefore, we concluded that playing stressful gameplay had no any impacts on both the brain and hormonal or biochemical properties in our proposed study.

3.2. During gameplay

We already found that mostly the pre-and post-condition did not affect the saliva amylase and showed less significant changes in the brain activity properties. For further investigation, we observed those activities while playing the game, separated into three segments (Game 1, Game 2, and Game 3). Figure 3 shows that the sAA’s average value decreased from 14.65 kU/l, 13.25 kU/l, and 12.25 kU/l during each segment. Even though there was no statistical difference between segments (p > 0.05), the tendency showed that the beginning of the assignment raised a higher possibility of mental stress occurrence.

From the perspective of brain waves activity, we initiated to observe the relative power during the games’ segments. Figure 4(a) shows that the band frequency between 8 and 15 Hz reduced during consecutive game play. The highest relative power was found during the first game with the relative power greater than 0.0001 compared to the other game trials (Game 2 and 3). Instead of observing the EEG's band powers on each electrode, we pictured the brain properties using a topographic map. The topographic map represented the band power ratio (specific absolute band power over the total band power) of each electrode. Figure 4(b) presents the grand average of the brain's relative band power after employing 30 secs epoch for 10 minutes during Game 1, Game 2, and Game 3. It can be seen that the temporal areas in the first game were also higher in the beta band than in the second and third games. It corroborated previous evidence that the onset of gameplay resulted in the increased physiological properties consistent with the

| Electrode Channels | Game 1 | Game 2 | Game 3 |
|--------------------|--------|--------|--------|
|                    | Mean   | SE     | Mean   | SE   | Mean   | SE   |
| Fp1-Av*            | 0.1977 | 0.0098 | 0.1896 | 0.0115 | 0.1895 | 0.0114 |
| Fp2-Av             | 0.2224 | 0.0131 | 0.2150 | 0.0155 | 0.2125 | 0.0158 |
| F7-Av*             | 0.3091 | 0.0097 | 0.2988 | 0.0099 | 0.2998 | 0.0106 |
| F3-Av              | 0.3405 | 0.0100 | 0.3356 | 0.0108 | 0.3393 | 0.0121 |
| Fz-Av              | 0.3001 | 0.0076 | 0.3002 | 0.0077 | 0.3009 | 0.0080 |
| F4-Av              | 0.3614 | 0.0093 | 0.3558 | 0.0106 | 0.3547 | 0.0111 |
| F8-Av**            | 0.3519 | 0.0141 | 0.3376 | 0.0149 | 0.3361 | 0.0142 |
| T3-Av*             | 0.5000 | 0.0239 | 0.4694 | 0.0259 | 0.4492 | 0.0238 |
| C3-Av*             | 0.3721 | 0.0145 | 0.3536 | 0.0147 | 0.3510 | 0.0131 |
| Cz-Av*             | 0.2840 | 0.0081 | 0.2777 | 0.0088 | 0.2760 | 0.0083 |
| C4-Av**            | 0.3513 | 0.0098 | 0.3356 | 0.0123 | 0.3337 | 0.0129 |
| T4-Av**            | 0.5092 | 0.0184 | 0.4822 | 0.0191 | 0.4700 | 0.0207 |
| T5-Av**            | 0.3527 | 0.0163 | 0.3361 | 0.0167 | 0.3317 | 0.0161 |
| P3-Av**            | 0.3182 | 0.0134 | 0.3054 | 0.0137 | 0.3031 | 0.0125 |
| Pz-Av**            | 0.2778 | 0.0099 | 0.2696 | 0.0106 | 0.2684 | 0.0103 |
| P4-Av**            | 0.3029 | 0.0115 | 0.2935 | 0.0122 | 0.2911 | 0.0119 |
| T6-Av*             | 0.3460 | 0.0163 | 0.3296 | 0.0161 | 0.3270 | 0.0163 |
| O1-Av              | 0.3825 | 0.0169 | 0.3629 | 0.0169 | 0.3614 | 0.0164 |
| O2-Av              | 0.3539 | 0.0160 | 0.3416 | 0.0160 | 0.3390 | 0.0158 |

Table 2. The beta-band relative power between different games’ segments (*p<0.05, **p<0.01)
biomedical process trend observed in salivary amylase activity.

Table 2 shows the relative beta band power on the site of the whole electrodes. Generally, the beta activity was reduced after Games 1, 2, and 3. However, we only found significant differences between the first (Game 1) and second segments (Game 2). The differences occurred on Fp1, F7, F8, T3, C3, Cz, C4, T4, T5, P3, Pz, P4, and T6. The most significant differences can be found in F8, C4, T4, and P4 (p < 0.01). Specifically, the most significant differences were found in the right hemisphere (F8, C4, T4, and P4; p < 0.01). Mostly, brain activity was affected in the temporal and parietal areas while playing the game.

The beta activity showed a predominant activity compared to other brain waves (delta, theta, and alpha). After the first segment (Game 1), the temporal sites was found higher than the other site. The statistical test demonstrated that the beta band

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Fig. 4. An example of relative brain wave changes on a subject during gameplay. (a) The brain waves activity on T3 site during Game 1, Game 2, and Game 3; (b) The topographic map of brain waves (delta, theta, beta, and alpha) during Game 1, Game 2, and Game 3.
power of the T4 site reduced from Game 1 (0.5092) to Game 2 (0.4822) and was significantly different (p < 0.01). However, it did not take place between the segment of Games 2 and 3.

3.3. During Pre-/Post Gameplay and Between Gameplays

To observe brain properties during pre-experienced or non-gameplays (before and after) and gameplays (Game 1, 2, and 3), we compared the circumstances of before – during gameplay and after – during gameplay. As indicated in tables 3, 4, 5, and 6, with a p-value less than 0.001, we discovered that the brain waves were significantly different in various situations, particularly in the delta, theta, and beta bands.

The most contributing band power was theta band (4-7 Hz), where almost all electrodes were higher during gameplay (game 1, 2, and 3) compared to the non-gameplays conditions (before and after gameplay) (table 4). The differences between before and after during gameplay were estimated to be between 0.10 to 0.15 relative powers, where the brain waves were more active during gameplay than during pre-experienced conditions. The significant electrodes were found on frontal (Fp1, Fp2, F7, F3, Fz, F4, F8), central (C3, Cz, C4), and parietal

| Electrode Channels | Before Game 1 | Game 2 | Game 3 | After |
|--------------------|----------------|--------|--------|-------|
| Fp1-Av             | 0.4915*±0.009  | 0.4440*| 0.4622*| 0.4718*| 0.1398 |
| Fp2-Av             | 0.4740±0.020   | 0.4089  | 0.4427  | 0.4550  | 0.1382 |
| F7-Av              | 0.4095±0.017   | 0.2714  | 0.2838  | 0.2818  | 0.1373 |
| F3-Av              | 0.3132±0.0168  | 0.2020  | 0.2109  | 0.2040  | 0.1274 |
| Fz-Av              | 0.2913±0.0120  | 0.1929  | 0.1962  | 0.1958  | 0.1285 |
| F4-Av              | 0.2945±0.0139  | 0.1875  | 0.1931  | 0.1988  | 0.1317 |
| F8-Av              | 0.3766±0.0144  | 0.2361  | 0.2557  | 0.2559  | 0.1312 |
| T3-Av              | 0.2991±0.0187  | 0.1231  | 0.1525  | 0.2069  | 0.1697 |
| C3-Av              | 0.2916±0.0204  | 0.1936  | 0.2151  | 0.2171  | 0.2782 |
| Cz-Av              | 0.3204±0.0179  | 0.2453  | 0.2605  | 0.2643  | 0.3189 |
| C4-Av              | 0.3062±0.0227  | 0.2034  | 0.2178  | 0.2221  | 0.2994 |
| T4-Av              | 0.3042±0.0227  | 0.1130  | 0.1346  | 0.1477  | 0.3034 |
| T5-Av              | 0.3338±0.0194  | 0.2260  | 0.2498  | 0.2586  | 0.3308 |
| P3-Av              | 0.3288±0.0219  | 0.2522  | 0.2682  | 0.2741  | 0.3273 |
| Pz-Av              | 0.3437±0.0201  | 0.2768  | 0.2909  | 0.2957  | 0.3458 |
| P4-Av              | 0.3350±0.0219  | 0.2491  | 0.2665  | 0.2700  | 0.3332 |
| T6-Av              | 0.3341±0.0203  | 0.2274  | 0.2521  | 0.2568  | 0.3322 |
| O1-Av              | 0.3293±0.0204  | 0.1908  | 0.2218  | 0.2324  | 0.3228 |
| O2-Av              | 0.3394±0.0188  | 0.2116  | 0.2357  | 0.2475  | 0.3260 |
lobes (P3, Pz, P4) and affected both in the right and left hemispheres.

Other significant differences could be observed in delta (0.5-4 Hz) and beta band powers (13-30 Hz) (tables 3 and 6). Since delta activities corresponded to the lower frequencies, it was evident that the relative band powers were lower during gameplay conditions than in non-gameplays conditions. It can mostly be found in Fp1, F7, F8, T3, and T4. Occipital lobes also had differences in O1 and O2 before and during the first gameplay (Game 1). Oppositely, the beta band (table 6) showed higher relative band powers during gameplays rather than in pre-experienced conditions (before and after) with 0.03 to 0.13 differences in relative band powers as found in F7, F8, T3, T4, T5, T6, and O1. However, only O2 showed differences before and during gameplay of 0.04-0.05 relative band powers differences.

The alpha band (8-12 Hz) was found not having significant differences compared to other band powers (table 5). F7 became the only electrode site in which the before gameplay had lower relative band powers compared to Game 1 and 2 conditions.

3.4. Intrasubject during gameplay

To enrich our findings, we also investigated the intrasubject during gameplay. On each subject, we observed how the brain changes during games 1, 2, and 3 and found any consistent pattern. Figure 5 and 6 show the T4 changes during gameplay from twenty subjects. From those figures, we recognized more
than 70% of subjects (15/20 subjects) had consistent patterns that tended to decrease (G1>G3) in the beta band across subjects (figure 6). It showed that in the first beginning of gameplay, the relative band powers were higher than in the last gameplay (game 3). This intrasubject analysis was similar to the topographic map properties in figure 4, where the Game 1 beta band power was also found higher in the temporal area compared to the last gameplay (Game 3). Oppositely, the subjects showed lower relative powers in the beginning compared to the last game (G1<G3) according to the delta band properties (figure 5). Despite other subjects depicting different trends, our data showed that brain activity in higher band frequency (beta band) tended to have higher band powers. It was opposite to the lower frequency (delta band) during the first gameplay experience (Game 1/G1).

3.5. Brain waves and salivary amylase activity changes

Nine out of twenty subjects had a salivary amylase activity greater than 30 kU/L. Thus, we examined whether there were significant changes in the brain waves associated with sAA-specific indexes of less than or equal to 30 kU/L and greater than that. Figure 5 demonstrates that beta-band power increased in the frontal, central, and parietal lobes in response to a higher sAA level (> 30 kU/l). In contrast, beta-band power decreased in response toward a lower sAA level (≤ 30 kU/l). Fz (p = 0.0015), F4 (p = 0.0030), Cz (p = 0.0069), C4 (p = 0.0079), T4 (p = 0.0063), Pz (p = 0.0046), P4 (p = 0.0020), T6 (p = 0.0059), and O1 (p = 0.0045) were the electrode sites that were clearly distinguishable. Likewise, the greatest difference was observed at the frontal Fz site (sAA > 30 kU/l, mean ± SE = 0.3288 ± 0.0074 vs sAA ≤ 30 kU/l, mean ± SE = 0.2953 ± 0.0043). Moreover, the right hemisphere (F4, C4, T4, P4, and T6) significantly contributed to more than the left hemisphere to distinguishing the two categories of sAA index levels. Finally, this result showed that higher beta band powers represented the higher stress level or frustration while playing a stressful game based on sAA measurement.
3.6. Discussion

Currently, a short time of stressor estimation is highly demanded; therefore, a proper assessment method is required. The sAA measurement is derived from the Hypothalamic-Pituitary-Adrenals (HPA) mechanism and could be a validator. Previous studies reported how brain activity represented the sAA assessment as a mental stress estimator [21], [32]. Al-Shargie et al. found that the right prefrontal cortex increased the ability of stress detection after receiving stress-induced by using the Montreal Imaging Stress Test (MIST). Their study showed that the reduced alpha band powers on the prefrontal were more accurately detecting the mental stress. However, their study found that the right hemisphere of the brain significantly changed during the second experiment after getting negative feedback. The right hemisphere tended to decrease and followed by the increase of the left hemisphere. Also, our proposed study confirmed that the right brain region became the most significant part that changed during mental stress changes. Besides, according to our experimental design, the alteration could be different compared to the previous studies on how the brain properties react. We assumed that this is also related to the environments that correlate the specific emotions and/or other psychological conditions of the participants.

The greatest anticipated change between segments (Game 1, 2, and 3) was on the right temporal region of the brain, according to our study. We hypothesized that this variation was determined by the game's audio. We suspected that this adjustment was influenced by the game's sound and the players' annoyance with hearing the same sounds multiple times to complete the objective. It conforms to the findings of Hébert et al., who revealed a temporal brain response to video game playing and investigated the role of in-game music. In addition, they discovered that the cortisol level increased in response to the mental stress modification [33].

Finally, we found that mental stress changes could be affected by computer gameplay, and this also corresponded to the previous study that investigated the emotional changes during playing games [34]. Predominantly, the brain's right hemisphere is also influenced by psychological changes and is also related to the fundamental study regarding the HPA axis. It could provide better mental stress detection accuracy [35], [36]. The large reviews on EEG utilization to detect mental health are still unable to find an agreement regarding superior band powers to locate stress conditions [19]. Past studies used Alpha band power to distinguish between stress and non-stress conditions. It also corresponded to the changes in the cortisol level [32]. Also, previous studies found the liability of beta band power following the statistical test showing that the reduced alpha band power was essential rather than the enhanced beta-band power. Our results showed that the beta band power was essential regarding the biochemical process alteration. It responded significantly to biochemical property changes represented by the sAA levels rather than the alpha band properties. From these findings, our study extended the previous studies that the sAA's values as a mental health objective measurement were affected by the environments and individual preferences in a specific condition. The prefrontal cortex does not always represent the stressors as previously reported. It might be due to the various activities that affect brain areas. Our results proposed that the right hemisphere should be investigated for further research together with numerous emotions and mental stress assessments.

Regarding specific index levels of sAA [12], we found that the EEG beta band power successfully reflected the two categories of sAA's levels. Higher salivary amylase activity increased the beta band activity. Previous studies reported that cortisol secretion is related to EEG wakefulness alertness in humans [37]. Another study reported that the increase of beta band power might reflect top-down attentional modulation after a stress-induced environment [38]. Therefore, we believed that the increased mental stress possibility enhances brain alertness as a consequence. Thus, further studies need to be employed to verify this finding.

4. Conclusion

Based upon our proposed study, we concluded that a stressful game can affect a brain waves activity. This study also found that the brain waves activity corresponded to the hormonal changes in salivary while playing a computer game. Predominantly, the beta brain waves in the central, temporal, and parietal lobes reflected the changes properly. This study confirmed that the topographic map of the temporal lobe in the right hemisphere probably played an essential part in the psychological changes. Therefore, this study can be an additional finding to extend the investigation of eustress-distress and other emotional changes caused by stress-induced environments. This result is evidence that stressful environments can simultaneously change hormonal and brain activity. Furthermore, future studies are urgently needed to investigate the cognitive functions that reflect psychological changes.

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