An Evaluation of Resting Tidal Volume Using a Biopac System Spirometer in Comparison to the Benchmark

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Introduction

Spirometers are devices that are commonly used to determine lung function by measuring tidal volume (TV) and airflow (Gildea et al. 2010). TV is the volume of air that enters and exits the lungs in a single inhalation and subsequent exhalation during any point of physical activity. Resting tidal volume (RTV), however, is a measurement of respiration that is collected when a person is breathing normally in a relaxed, homeostatic state (Widmaier et al. 2016). Thus, measurements of RTV are typically lower than total TV because total TV is subject to fluctuations during periods of physical activity when the body requires more oxygen. Comparisons in TV are made in relation to the accepted standard value for RTV in human adults, 500 milliliters (mL), depending on body size (Widmaier et al. 2016).

Though spirometers are commonly used in a clinical setting to support diagnosis of respiratory diseases (Gildea et al. 2010), this paper addresses the use of spirometers in conducting research studies. A previous experiment at the University of Wisconsin-Madison was conducted on human subjects to test the ability of water-filled spirometers to measure RTVs that were close to the benchmark value of 500 mL. Average RTV was calculated for 181 male and 267 female participants and determined to be 735.2 mL with a standard deviation of 365.2 mL and 609.3 mL with a standard deviation of 302.4 mL, respectively (courtesy of Dr. Andrew Lokuta, *unpublished data*). The results of this study suggested a large deviation from the accepted RTV of 500 mL and variability among participants using the water-filled spirometer. One suggested explanation may be that the water-filled spirometer created a physical resistance against the participant's airflow due to the unequal pressure within the water-filled device compared to the air pressure of the testing environment, resulting in more forceful respirations (Mottram, 2018, *in personal communication with Dr. Lokuta*). Another explanation could be that the participants were aware their respiration was being tested, causing them to consciously control their breathing. In modification of this previous experiment, the following study was conducted to measure RTV in human subjects using a Biopac spirometer and an experimental design that presented the participants with a distraction to minimize conscious respiratory control. This spirometer was utilized in this comparative study as a water-filled spirometer was unavailable for experimentation. Additionally, the Biopac spirometer is an improvement from the water-filled spirometer because the device has an aperture that allows for equal pressure within the device compared to the air pressure in the testing environment, thus allowing for less resistance to airflow (Rebuck et al. 1996, *in personal communication with Dr. Andrew Lokuta*).

An examination of the Hawthorne effect prompted the decision to include a distraction in this experimental design. This effect states that expected outcomes of an experiment can change depending on an individual's knowledge of the variable of interest (McCarney et al. 2007). Many previous studies have indicated that intrusive techniques, such as the utilization of spirometers, focus an individual's attention on their breathing. This behavioral control has led to significant effects on breathing patterns (Etzel, 2006).

Therefore, in regard to the following study examining RTV, it was assumed that participants were aware that their breathing was being monitored because they were interacting with devices used to measure their respiration. Thus, it was expected that this awareness would result in conscious control of breathing and abnormal fluctuations in RTV, which could result in deviations from the benchmark RTV of 500 mL. To increase the likelihood for spontaneous, resting-state breathing, studies have suggested the use of task engagement or distraction (Boiten, 1998). Hence, the following study utilized an auditory and visual distraction to divert the participant's focus from controlling their breathing in order to reduce the Hawthorne effect.

In the following study, it was hypothesized that the use of an auditory and visual distraction would reduce the effect of confounding variables on the data, resulting in a RTV that more closely reflects the benchmark value of 500 mL. The ability to observe a RTV that is closer to this value than in previous experimentation using the water-filled spirometer would suggest that the Biopac System spirometer better reflects the benchmark for measuring RTV. This hypothesis was tested by studying three physiological measurements: TV, skin conductance and respiration rate (RR).

TV was the main physiological measurement that was evaluated in this study, whereas RR and skin conductance were supporting variables that could provide an explanation for large deviations in RTV. These measurements were deemed important because variations in respiration reflect changes in mood, which is also a causal factor in skin conductance changes (Etzel, 2006). For example, mood disorders, such as anxiety, have been associated with hyperventilation and increased levels of electrodermal activity (EDA) (Wilhelm and Roth, 2001). Sweat contains saltwater which conducts electricity well, so stimulation of sympathetic nervous system activity in mood disorders causes increased sweat secretion, resulting in higher EDA values, as well as altered respiration patterns (Etzel, 2006). Thus, the relationship between skin conductance and respiration measures was utilized in the following study.

Measuring these supporting variables throughout both the baseline and distraction periods was done to examine how a distraction period could potentially influence skin conductance and RR, resulting in a more relaxed state. Furthermore, the results of this study allow for a comparison of the Biopac System spirometer in relation to the benchmark, which will be helpful information for future studies using this Biopac technology.

Materials

Tidal volume (TV), respiration rate (RR), and skin conductance were all examined using three different measurement devices. A spirometer and its separate 2.0 L calibration syringe (Model: SS11LA, SN: 12128333, Biopac Systems, Inc. Goleta, CA) were used for calibration and measurement of TV in milliliters (mL). An additional supply of mouth pieces, air filters and nose clamps were utilized for sanitary purposes and accurate TV measurements through mouth exhalation. RR, in breaths per minute (BPM), was studied using a respiratory belt (Model: SS5LB, SN: 1602007558, Biopac Systems, Inc. Goleta, CA). BSL EDA finger electrodes were used in conjunction with a Xdcr lead set (Model: SS3LA, SN: 11053677, Biopac Systems, Inc. Goleta, CA) to measure skin conductance in microsiemens (µS). Data recording and analysis was conducted using the Biopac Student Lab System (BSL 4 software, MP36) as well as consultation from the Biopac Systems, Inc. Student Manual (Biopac Systems Inc. ISO 9001:2008) for equipment setup.

A video was presented for the distraction period of the experiment. The *Small Thing Big Idea* video, "Why the Pencil is Perfect," was played in order to observe any measurable differences in TV that occurred upon distraction by this auditory and visual stimulus. This distraction was presented to divert the participants' attention away from the physiological measurements being collected to determine if spontaneous breathing could be induced, resulting in data reflecting the accepted literature TV value.

Methods

Participants

Participants, ages 20-25 were recruited on a voluntary basis from the University of Wisconsin-Madison. Physiological measurements were collected at the UW-Madison Medical Sciences Center. A consent form was signed by all participants describing the confidentiality measures utilized in the study and alerting them of any potential discomfort.

Procedure

Participants were eligible to participate in this study if they met the following criteria. The inclusion criteria included the willingness and ability to consent as well as falling into the age range of 20 to 25 years. The exclusion criteria of participants were determined based on the questionnaire the participants filled out following experimentation. If they indicated that they smoked, the data was excluded from the results. Participants with asthma were still included as they were not required to participate in physical activity nor were they having an asthma attack during the experiment.

The following Biopac System preparations were made prior to testing. The Biopac equipment – spirometer, respiration belt, and BSL EDA finger electrodes – was attached to the Biopac Student Lab System via channels 1, 2 and 3 respectively. “Airflow” (SSL11LA, SN) was selected for TV, measured in liters (L), on channel 1, “Respiration” (SS5LB) was selected for RR, measured in millivolts (mV), using channel 2, and “Electrodermal activity” (EDA, SS3L, SS3LA, SS57L, 0-35 Hz) was selected for skin conductance, measured in microsiemens (μS), on channel 3.

A respiratory belt was secured on each participant’s upper chest with the monitor centered on the sternum. The BSL EDA Finger Electrodes were strapped to the participant's index and middle fingers on their right hand after applying electrode gel. The spirometer was calibrated using the Biopac spirometer 2.0 L calibration syringe. Five full pumps of the syringe were recorded, and the graph was scanned for distinct peaks indicating the plunger’s movement to confirm that the system and spirometer were working properly. A disposable mouthpiece and air filter were attached, and the spirometer was given to the participant’s free hand. Each participant received a new mouthpiece and air filter in order to maintain consistency between trials and prevent transfer of bacteria between participants. Calibration procedures were conducted for the respiratory belt and BSL EDA Finger Electrodes as indicated by the Biopac Systems Inc. Student Manual. After the participants were instructed to take three deep breaths, the graphs were reviewed to identify peaks and troughs indicating a properly functioning Biopac system.

Experimentation was conducted in a private room to ensure minimal external disruptions that could alter the participant’s data. Two investigators stayed with the participant during the entirety of the study in order to verify that the measurements were continually recorded and to stop the trial if the participant felt uncomfortable at any time. The investigators were positioned out of the participant's view in order to reduce the risk of introducing a confounding variable that could influence the data measurements. The participant was instructed to sit with both feet on the floor facing the monitor. Data collection began at the start of a timed PowerPoint presentation that the participant was told to watch for the duration of the experiment. The experimental timeline included two baseline periods separated by a distraction period (Figure 1). An initial one-minute baseline recording of TV, skin conductance, and RR occurred while a blank PowerPoint slide was displayed. The "Why the Pencil is Perfect" video played for a duration of 3 minutes and 37 seconds and was followed by another minute of a blank PowerPoint slide. The video served as a distraction for the participants to limit voluntary control of breathing, resulting in what was expected to be a more accurate resting-state value for TV.

Upon completion, participants were assisted by an investigator to remove all Biopac devices and were told experimentation had concluded. A post-trial questionnaire asking participants to indicate their sex, asthma and smoking history, and height was collected. These details were reviewed against the data collected in order to detect potential confounding variables which may have provided reasoning for any outlier data observed. In addition, these factors could alter the participant's normal RTV, deviating from the accepted value of 500 mL.

Data Analysis

The average TV (mL) for each participant was measured before, during, and after the distraction period (Figure 1) using the spirometer. TV was determined by collecting the "P-P" (peak to peak) values of each inhalation and exhalation waveform, as calculated by the Biopac software, and dividing this number by two (Figure 2). The TV values for each breath taken were averaged to determine the individual's overall TV average throughout the first baseline period. This was repeated with the distraction period and the second baseline period. To determine the average TV of the entire sample for each period, a weighted average of the participants' individual averages was calculated.

Supplemental RR and skin conductance data was also collected using the Biopac program. RR (BPM) was determined by counting the number of peak-trough pairs in each participant's trial and dividing this value by the respective amount of time (in minutes) for each period. Sample averages for each period were also recorded. The average skin conductance for each participant was measured before, during, and after the distraction period, and a weighted average of the entire sample was calculated. The data collected from these calculations provided insight into how the participant's physiological responses reflected resting-state TV.

A one-sample, two-tailed t-test was conducted to compare the average TV data to the published RTV value of 500 mL. This type of statistical analysis was used to account for the potential for the sample mean to be greater than or less than the stated population mean. Multiple two-tailed t-tests were conducted. For each physiological measurement, the participant averages for the first baseline period were compared to those of the distraction period. Likewise, similar t-tests were conducted between the distraction period and the second baseline period. These statistical analyses were utilized to identify if the distraction period had an effect on the physiological measures. A final t-test was performed to compare average TV during the distraction period according to biological sex. This statistical value allowed for analysis of potential inherent differences in TV. If $p < 0.05$ the results were determined to be statistically significant. Skin conductance and respiration data were used to observe any potential correlation between variables or attempt to explain any deviations. These data allowed for comparisons with the TV data.

Positive Controls

To ensure functionality of the Biopac equipment, positive control tests were conducted by the investigators. Baseline measurements for TV, RR and skin conductance were taken for 2 minutes. These resting averages were found to be 480 ± 117.38 mL, 12.48 ± 4.10 BPM, and 2.18 ± 1.84 μS, respectively (n=2). Another set of measurements for TV, RR and EDA were taken following 2 minutes of running in place. These averages were found to be 851.50 ± 243.95 mL, 13.53 ± 3.56 BPM, and 2.87 ± 2.54 μS, respectively (n=2). All variable measurements were observed to increase as expected after a short period of activity. This indicated that equipment was functional and that measurements tested in this experiment exist.

Negative Controls

The initial and final minute of experimentation for each participant functioned as the negative control for their own data due to anatomical and physiological differences. These were baseline measurements without visual stimuli or experimental manipulations. Physiological changes observed during the distraction period of the experiment were compared against the two baseline periods to understand how the presentation of a distraction potentially affected participants' respiration. Any discrepancies between these periods allowed for analysis of the impact of video distraction on the physiological measurements studied in this experiment.

Results

Data was recorded from 30 participants, and all recorded participant data was included in the analysis as zero participants met the exclusion criteria. This experimental sample contained 13 male and 17 female subjects. All TV data was collected in the following order and reported as such; first baseline period, distraction period, and second baseline period. The average TVs were calculated and found to be 509.60 ± 160.39 mL, 489 ± 117.54 mL, and 501.37 ± 123.83 mL, respectively (n=30) (Figure 3). The ranges of average TV for the entire sample population were computed to be 570 mL, 510 mL, and 500 mL (Figure 4). Average TV was also separately calculated based on the participants' self-reported biological sex. The male participants' averages were found to be 529.92 ± 157.95 mL, 519.23 ± 140.15 mL, and 538.54 ± 142.46 mL (n=13). The female participants' averages were found to be 494.06 ± 165.30 mL, 465.88 ± 94.87 mL, and 472.94 ± 102.92 mL (n=17). Using the entire sample's average TVs, a two-tailed t-test was calculated between the first baseline period and distraction period and found to be 0.57. Likewise, the two-tailed t-test between the distraction period and the second baseline period was found to be 0.69. A one-sample, two-tailed t-test was conducted using each experimental period's average TV in comparison to the benchmark of 500 mL. P-values were determined to be 0.75, 0.61, 0.95. A final t-test was conducted between the female (n=17) and male (n=13) average TVs in the distraction period. The p-value was determined to be 0.23.

Calculations for EDA data were similarly conducted. All EDA data was collected in the following order and reported as such; first baseline period, distraction period, and second baseline period. Average EDA for the entire data set was found to be 5.08 ± 2.65 µS, 4.20 ± 2.08 µS, and 3.77 ± 2.05 µS, respectively (n=30) (Figure 5). Specifically, males were found to have average EDA values of 5.08 ± 2.73 µS, 4.03 ± 2.19 µS, and 4.09 ± 1.87 µS (n=13). Females, on the other hand, were found to have average EDA values of 5.10 ± 2.59 µS, 4.33 ± 2.00 µS, and 3.53 ± 2.14 µS (n=17). The ranges of average EDA values for the entire sample group were computed to be 11.63 µS, 11.25 µS, and 7.79 µS (n=30) (Figure 6). A two-sample t-test was conducted for the EDA sample averages between the first baseline period and the distraction period, resulting in a p-value of 0.15. The same test was run for the distraction period and second baseline period, resulting in a p-value of 0.42. EDA data was compared to TV data using correlation statistics, where the r-value was determined to be 0.05 (n=30).

All RR data was collected in the following order and reported as such; first baseline period, distraction period, and second baseline period. Average RRs for the entire sample were found to be 12.81 ± 3.34 BPM, 12.80 ± 3.40 BPM, and 10.69 ± 3.13 BPM, respectively (n=27) (Figure 7). Average RRs for males were calculated as 12.77 ± 3.37 BPM, 12.62 ± 3.54 BPM, and 10.67 ± 3.10 BPM (n=13). Furthermore, average RRs for females were calculated, finding 12.86 ± 3.45 BPM, 12.97 ± 3.38 BPM, and 10.71 ± 3.28 BPM (n=14). The range of average RR values for the entire sample group was computed to be 12.58 BPM, 14.27 BPM, and 11.24 BPM (n=27) (Figure 8). A two-tailed t-test was calculated between the first baseline period and distraction period and found to be 0.94. Likewise, the two-tailed t-test between the distraction period and the second baseline period was found to be 0.026. RR was determined to have an overall correlation coefficient of 0.11 with the TV data (n=27). A final correlation was conducted comparing EDA to RR (n=27), resulting in an r-value of 0.09.

Discussion

The results from the two-tailed t-test in this experiment (H_0: $\mu_{first\ baseline} = \mu_{distraction}$ H_a: $\mu_{first\ baseline} \neq \mu_{distraction}$, H_0: $\mu_{second\ baseline} = \mu_{distraction}$ H_a: $\mu_{second\ baseline} \neq \mu_{distraction}$) indicated that there was no statistical difference between the average TV of the baseline period and the distraction period TV ($p > 0.05$). Therefore, the hypothesis that a distraction period would lower TV by mitigating the Hawthorne effect was unsupported. The distraction period had no effect on TV, and benchmark values (H_0: $\mu_{first\ baseline} = \mu_{benchmark}$ H_a: $\mu_{first\ baseline} \neq \mu_{benchmark}$, H_0: $\mu_{distraction} = \mu_{benchmark}$ H_a: $\mu_{distraction} \neq \mu_{benchmark}$, H_0: $\mu_{second\ baseline} = \mu_{benchmark}$ H_a: $\mu_{second\ baseline} \neq \mu_{benchmark}$) were reflected in all three experimental periods ($p > 0.05$). Based on these results, a distraction period was not necessary to reach a TV of 500 mL.

This suggests that the Biopac System is more representative of the benchmark than the water spirometer (courtesy of Dr. Andrew Lokuta, *unpublished data*). There were lower average TVs reported using the Biopac spirometer regardless of the distraction period than with the water spirometer alone. However, due to limited knowledge about the past water spirometry participants, it was unknown if this experiment used a representative sample for accurate comparison between these studies. Characteristics such as physical activity level and height could vary between experimental samples, leading to different TVs.

In the current experiment using Biopac, height and TV were positively correlated, having a correlation coefficient of 0.63 (Figure 9). This was expected as greater height was positively correlated with larger lung size and capacity, therefore, a larger TV (Bhatti, 2014). This suggests that body size may play a role in altering TV. In each experimental period, the large TV range was likely affected, in part, by the participant height variability.

According to previous studies, a statistically significant relationship between TV, RR and EDA was anticipated in this experiment (Wilhelm and Roth, 2001). However, no correlation was found between EDA and TV, RR and TV, or EDA and RR. This unexpected finding could be the result of several possibilities. For example, this experiment consisted of a small sample size, 30 participants, who were enrolled in the same college course. This was not a representative sample for the entire population of 20-25 year olds and was therefore, biased. There were not enough participants in the sample to minimize standard error, meaning these results contain potentially misleading, atypical data. Additionally, this could be due to possible equipment malfunctions and human error, such as an unsecure respiratory belt or an inadequate amount of gel for the EDA electrodes.

Further statistical testing indicated that there was no statistical difference in EDA values between any of the experimental periods ($p > 0.05$). This means that the distraction had no effect on EDA. Likewise, RR between the first baseline period and the distraction period was found to have no statistical difference ($p > 0.05$). A statistical difference was found, however, in RR between the distraction period and the second baseline period ($p < 0.05$). This was likely due to participants' adjustment to the experimental design and apparatus as the first and second baseline periods utilized identical blank PowerPoint slides. While this could be a direct result of the distraction period, it's unlikely because there was no statistical difference found among any other physiological measurements.

Conclusions were also made indicating that, based on this data, there was no difference in TV based on biological sex. A two-tailed t-test (H_0: $\mu_{male} = \mu_{female}$ H_a: $\mu_{male} \neq \mu_{female}$) was conducted, indicating that there was no significant difference between male and female TV during the distraction period ($p > 0.05$).

Following experimentation, possible limitations were addressed in relation to the experimental results. Systematic errors, such as selection bias and confirmation bias, likely occurred throughout the study. The participants were not randomly selected to participate in this study, but instead, were selected from a specific college course, skewing the data. Also, it was assumed that the investigators, knowing the hypothesis in question, subconsciously affected the data collection process, altering it in favor of the anticipated outcome.

The data illustrated a large range of TVs with high standard deviations for each experimental period. Tidal volume values were expected to be higher than the benchmark of 500 mL, but TVs as low as 300 mL were recorded. This does not necessarily suggest errors in data collection but may simply reflect the accuracy of the Biopac spirometer in measuring the benchmark. The large ranges in TV can also be attributed to the physiological differences between participants.

The intrusive nature of the spirometer also likely altered the observed results slightly. Previous papers have discussed the limitations of spirometry due to psychological awareness of the respiration-measuring instrument, causing disruptions in normal respiratory patterns. While there are measurement tools that are less invasive, they are less accurate as a slight movement can require recalibration (Etzel, 2006). However, based on the results using the Biopac spirometer, this psychological awareness was not particularly evident as a distraction was unnecessary to achieve the benchmark.

In future studies, it would be advantageous to modify the experimental design and test a larger sample of participants. Given that the data did not support the effectiveness of a video distraction in decreasing TV, there is no advantage to including this in future experimental designs. However, the lack of statistical difference in TV between experimental periods may have been due to the small sample size (n=30) tested in this study. There were likely inconsistencies in relation to the entire population of 20-25 year olds. The data reflects the demographic of a small portion of college students attending the University of Wisconsin-Madison. This indicates that the conclusions made in this study may not be true outside of this experimental sample. Also, the sample in this experiment is not exchangeable with that of the study determining the benchmark RTV. There are many variables, including age, height, and personal health histories that could drastically alter this RTV comparison. Therefore, future experimentation should include a larger number of participants, random selection and more demographic diversity to reflect the overall population. This will allow for results that more closely reflect the true population value. Strongly recommend more aggressive recruiting

Vital capacity is a measurement of interest in future studies as it lends further insight into physiological differences among participants. Average vital capacity measurements will be useful in order to compare a participant's tidal volume data to their inspiratory and expiratory reserve volumes as well as residual volume. Therefore, it is strongly encouraged that future studies utilize forced vital capacity maneuvers and standardize each participant's data to their chest volume and dimensions. This will provide an individualized normalization tool in order to better understand if the tidal volume differences seen are indicative of participant variation or experimental manipulation.

The results of this study indicated that the benchmark can be achieved using the Biopac spirometer; however, there was a large amount of variance in average TV during each experimental period. Therefore, future studies should focus on decreasing the variance across participants through meticulous data collection. Testing a larger sample size would also likely decrease the large standard deviations seen in this study.

Conclusion

In conclusion, this data shows that the Biopac spirometer is a more accurate tool to measure benchmark RTV than the water spirometer. The participants were able to reach a RTV of 500 mL using the Biopac spirometer, even without a distraction. Further, the experimental periods had no effect on any of the physiological measures tested except for RR. The average RRs between the distraction period and the second baseline period were determined to be statistically significant. This indicates that the video either had an effect on the participants' respirations immediately following the video or that the participants became accustomed to the experimental environment. Future studies are required to determine which explanation applies to this observed outcome. In addition, height and TV had a positive correlation, indicating that controlling for body size in future experiments may be advantageous.

Figures

Figure 1: Experiment Timeline. The participant was connected to the Biopac spirometer, respiration belt, and BSL EDA finger electrodes to measure physiological responses throughout the entire experiment while continuously recording measurements. The study was started with a calibration period (< 1 minute) to ensure that the equipment was correctly fitted to the participant and all devices were accurately recording data. The first baseline period (1 minute) was characterized by the physiological measures of the participant without visual or auditory stimuli. This acted as the negative control because it measured each participant's RR, TV and skin conductance without any environmental manipulations. Next, the distraction period (3 minutes, 37 seconds) was presented to each participant with both visual and auditory stimuli from a video playing on the computer screen in front of them. Finally, the second baseline period (1 minute) was comprised of the participant's physiological response to another period without visual or auditory stimuli. Again, this baseline period acted as a negative control, and the data collected was compared to the first baseline period and distraction period.

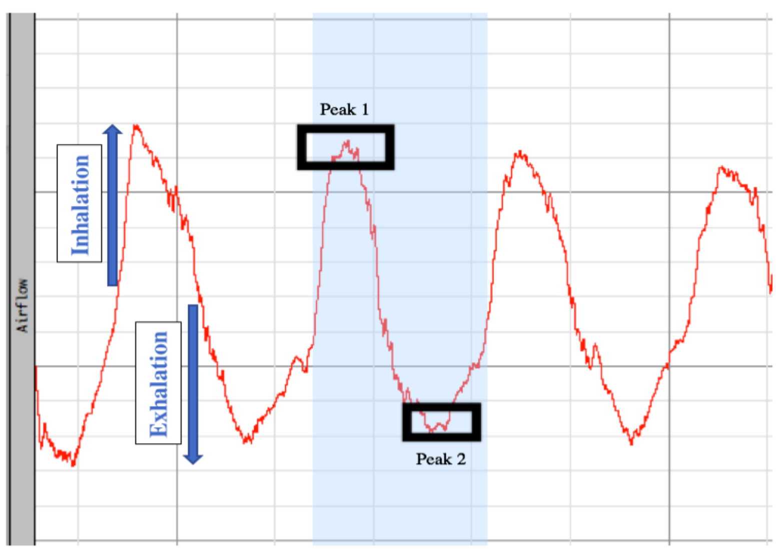

Figure 2: Illustration of Tidal Volume Data Analysis. An illustration of the raw data collected from a TV analysis using the Biopac system. The x-axis was measured in units of time (seconds) and the y-axis was measured in units of airflow (mL). TV was determined by measuring the "P-P" (peak-to-peak) values of each inhalation and exhalation waveform and dividing this number in half. All of the "P-P" values for each respiratory cycle were then averaged to determine the individual's overall average TV during the experimental period.

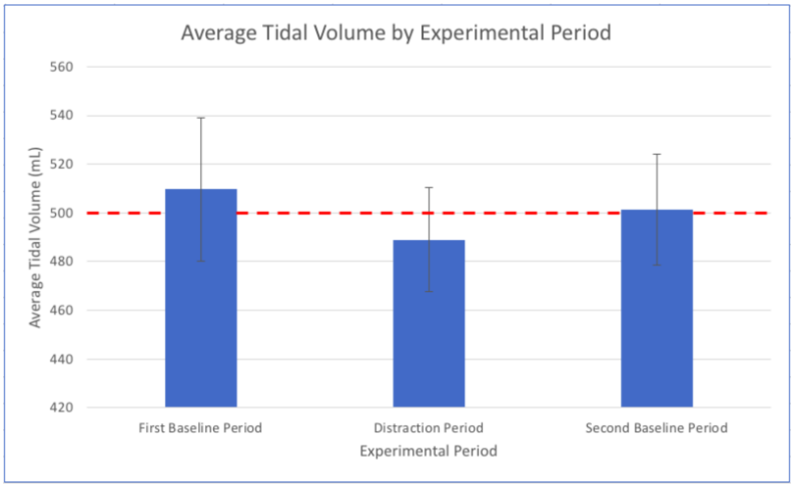

Figure 3: Average Tidal Volume by Experimental Period. The average TV for each participant was used to find the overall average TV of participants (n=30) in each experimental period. The average TV was 509.60 mL ($\sigma_{\bar{x}}$= 29.28) for the First Baseline Period, 489.00 mL ($\sigma_{\bar{x}}$ = 21.46) for the Distraction Period, and 501.37 mL ($\sigma_{\bar{x}}$ = 22.61) for the Second Baseline Period. The standard error for each experimental period was indicated using error bars. A decrease in TV was observed from the First Baseline Period to the Distraction Period, followed by an increase in TV from the Distraction Period to the Second Baseline Period. No statistical significance was found between the average TVs of each experimental period ($p > 0.05$). The benchmark was indicated using a red dashed line, illustrating the comparison of the average TV for each experimental period in relation to the benchmark of 500 mL ($p > 0.05$).

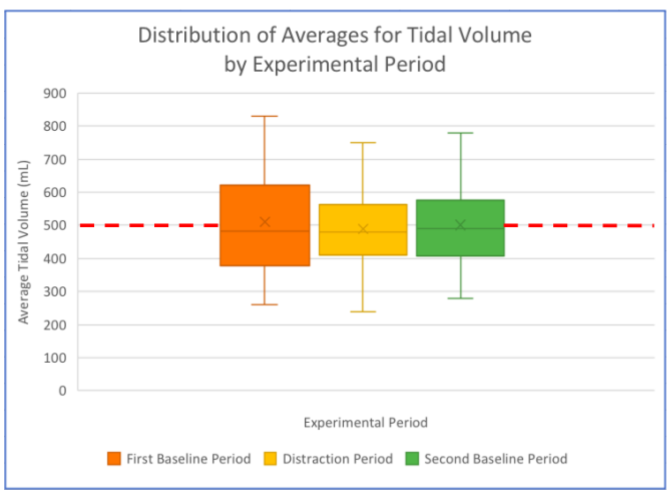

Figure 4: Distribution of Averages for Tidal Volume by Experimental Period. The distribution of average TV for each participant (n=30) was illustrated using box plots according to experimental period. For the First Baseline Period, the median value was 482.5 mL, the upper and lower quartiles were 622.5 mL and 377.5 mL with an interquartile range of 245 mL. The highest and lowest observations were 830 mL and 260 mL with a range of 570 mL. For the Distraction Period, the median value was 480.0 mL, the upper and lower quartiles were 562.5 mL and 410.0 mL with an interquartile range of 152.5 mL. The highest and lowest observations were 750 mL and 240 mL with a range of 510 mL. For the Second

Baseline Period, the median value was 490.0 mL, the upper and lower quartiles were 576.75 mL and 407.5 mL with an interquartile range of 162.25 mL. The highest and lowest observations were 780 mL and 280 mL with a range of 500 mL. The benchmark was indicated using a red dashed line, illustrating the comparison of the average TV for each experimental period in relation to the benchmark of 500 mL. No statistical significance was observed between the distributions of average EDA for each experimental period ($p > 0.05$).

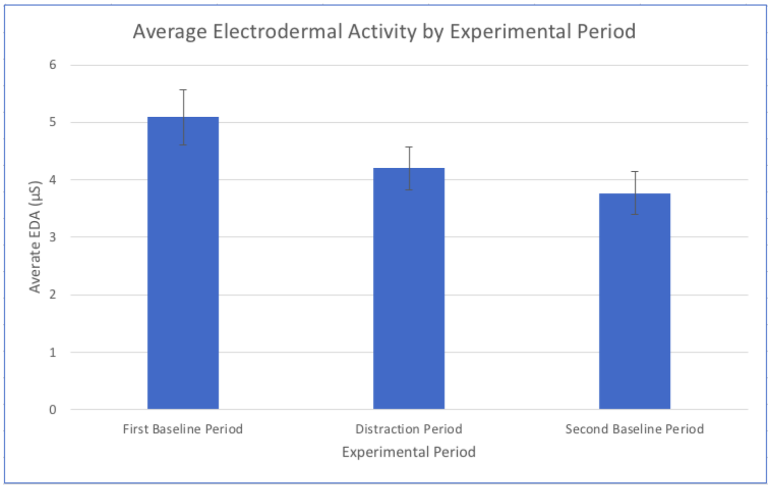

Figure 5: Average Electrodermal Activity by Experimental Period. The average EDA for each participant was used to find the overall average EDA of participants (n=30) in each experimental period. The average EDA was 5.08 µS ($\sigma_{\bar{x}}$ = 0.48) for the First Baseline Period, 4.20 µS ($\sigma_{\bar{x}}$ = 0.38) for the Distraction Period, and 3.77 µS ($\sigma_{\bar{x}}$ = 0.37) for the Second Baseline Period. The standard error for each experimental period was indicated using error bars. A decrease in EDA was observed from the First Baseline Period to the Distraction Period, followed by another decrease in EDA from the Distraction Period to the Second Baseline Period. No statistical significance was found in the average EDAs between the First Baseline Period and the Distraction Period as well as the Distraction Period and the Second Baseline Period ($p > 0.05$).

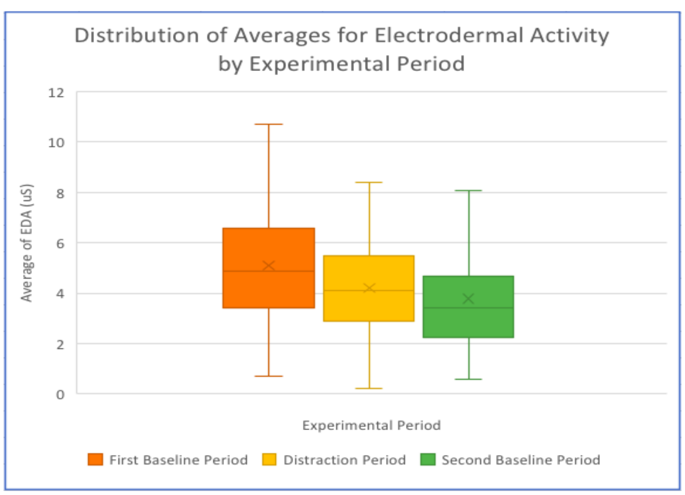

Figure 6: Distribution of Averages for Electrodermal Activity by Experimental Period. The distribution of average EDA for each participant (n=30) was illustrated using box plots according to experimental period. For the First Baseline Period, the median value was 4.86 µS, the upper and lower quartiles were 6.56 µS and 3.42 µS with an interquartile range of 3.14 µS. The highest and lowest observations were 10.72 µS and 0.69 µS with a range of 10.03 µS. For the Distraction Period, the median value was 4.12 µS, the upper and lower quartiles were 5.47 µS and 2.87 µS with an interquartile range of 2.6 µS. The highest and lowest observations were 8.4 µS and 0.22 µS with a range of 8.18 µS. For the Second Baseline Period, the median value was 3.41 µS, the upper and lower quartiles were 4.67 µS and 2.26 with an interquartile range of 2.41 µS. The highest and lowest observations were 8.09 µS and 0.58 µS with a range of 7.51 µS. No statistical significance was observed between the distributions of average EDA for each experimental period ($p > 0.05$).

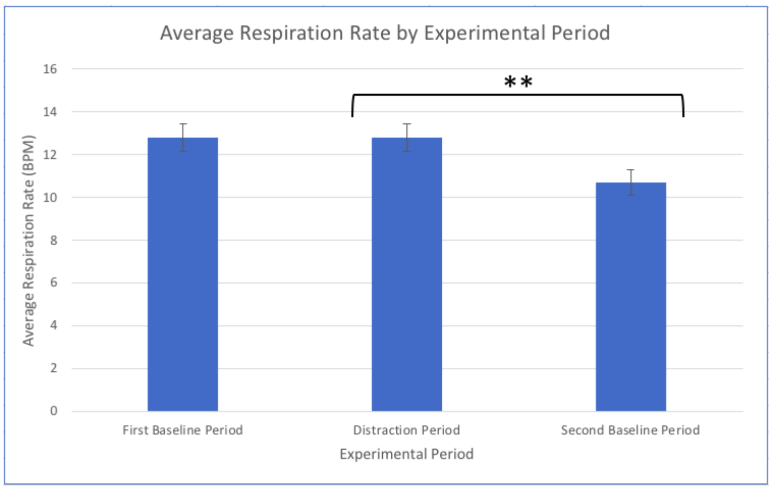

Figure 7: Average Respiration Rate by Experimental Period. The average Respiration Rate (RR) for each participant was used to find the overall average RR of participants (n=27) in each experimental period. The average RR was 12.81 BPM ($\sigma_{\bar{x}}$= 0.66) for the First Baseline Period, 12.80 BPM ($\sigma_{\bar{x}}$ = 0.67) for the Distraction Period, and 10.69 BPM ($\sigma_{\bar{x}}$ = 0.61) for the Second Baseline Period. The standard error for each experimental period was indicated using error bars. A slight decrease in RR was observed from the First Baseline Period to the Distraction Period, followed by another decrease in RR from the Distraction Period to the Second Baseline Period. No statistical significance was observed between the First Baseline Period and Distraction Period. Statistical significance was found between the Distraction Period and Second Baseline Period ($p < 0.05$), indicated by (**).

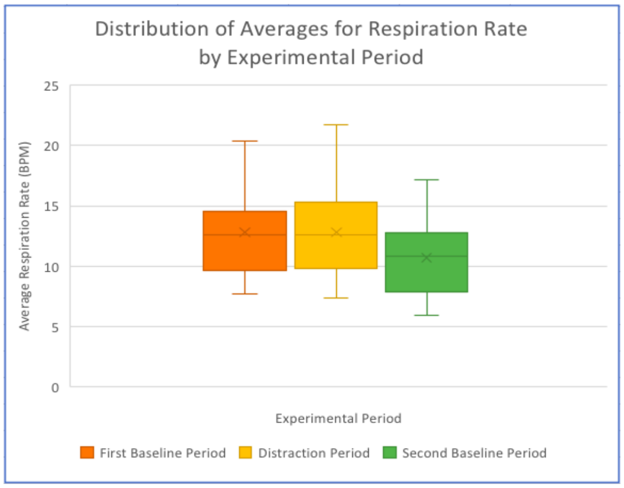

Figure 8: Distribution of Averages for RR by Experimental Period. The distribution of average Respiration Rate (RR) for each participant (n=27) was illustrated using box plots according to experimental period. For the First Baseline Period, the median value was 12.58 BPM, the upper and lower quartiles were 14.52 BPM and 9.68 BPM with an interquartile range of 4.84 BPM. The highest and lowest observations were 20.32 BPM and 7.74 BPM with a range of 12.58 BPM. For the Distraction Period, the median value was 12.56 BPM, the upper and lower quartiles were 15.33 BPM and 9.78 BPM with an interquartile range of 5.55 BPM. The highest and lowest observations were 21.67 BPM and 7.40 BPM with a range of 14.27 BPM. For the Second Baseline Period, the median value was 12.38 BPM, the upper and lower quartiles were 12.79 BPM and 7.87 BPM with an interquartile range of 4.92 BPM. The highest and lowest observations were 17.14 BPM and 5.90 BPM with a range of 11.24 BPM. No statistical significance was observed between the distributions of average RR for each experimental period ($p > 0.05$).

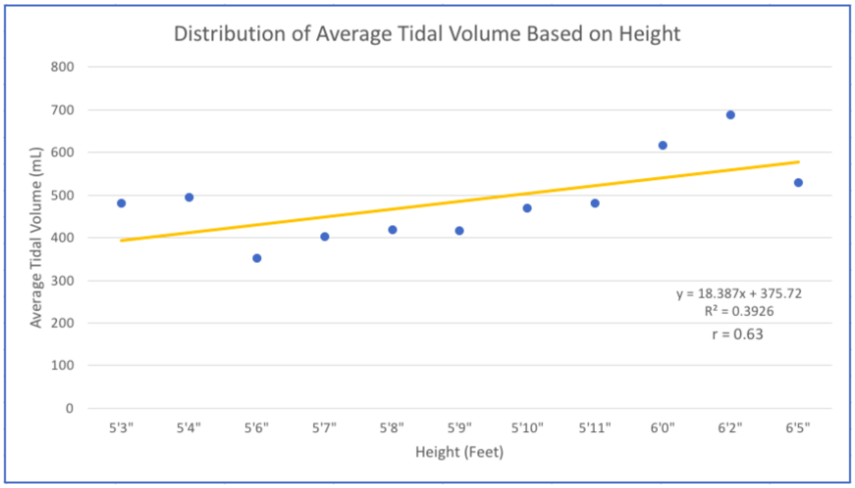

Figure 9: Distribution of Average Tidal Volume Based on Height. Tidal Volume (TV) for each participant was graphed in relation to self-reported height (n=25) to determine correlation. Any duplications in height value were averaged to function as a single point on the graph. A line of best fit was added to reflect the correlation between TV and height. Correlation statistics reported a correlation coefficient of 0.63, which indicates a positive correlation between the two variables.

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Link to the video used in Distraction Period:

https://www.facebook.com/SmallThingBigIdea/videos/1344073369072021/