

NEUROSTAT

TUTORIAL and MANUAL

Version 2.0

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Introduction:

NeuroStat is a program that provides statistical comparisons and descriptive statistics of EEG samples saved as Individual NeuroGuide Analysis Files or *.NGA files.

NeuroStat also provides individual and group parametric statistical tests using the Key Institute LORETA program. Individual comparison options are independent t-tests, paired t-tests and ANOVA as well as absolute and percent differences. Individual statistics are used for pre vs post treatment comparisons or differences between eyes closed and eyes open, etc.

Group comparative statistics may require the use of [NeuroBatch](#) to produce a NeuroGuide Group Analysis File or *.NGG file of the surface EEG. Group paired t-tests do not require the use of NeuroBatch and LORETA statistical analyses do not require the use of NeuroBatch. Descriptive Group statistics include Means and Standard Deviations and independent t-tests between Groups (does not assume equal variance or sample sizes). Group statistics are used to compare different groups of subjects such as 100 ADD children to 100 normal children, etc. Use [NeuroBatch](#) to create the *.NGG files before using NeuroStat.

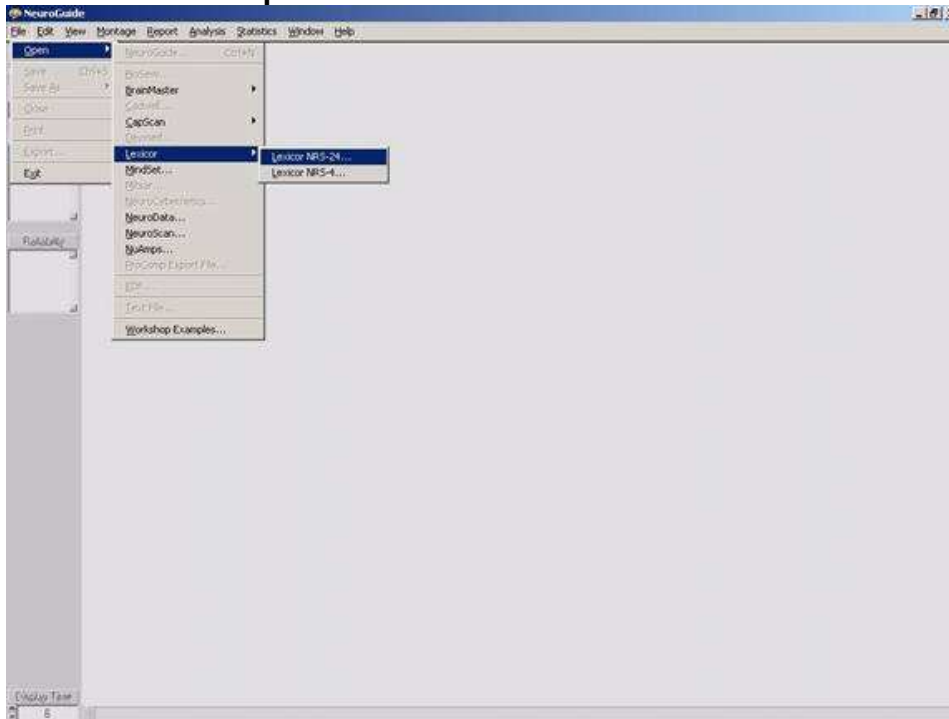
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Step #1: In Demo Mode Hold the Left Mouse Button and Click File>Open>Lexicor NRS24.

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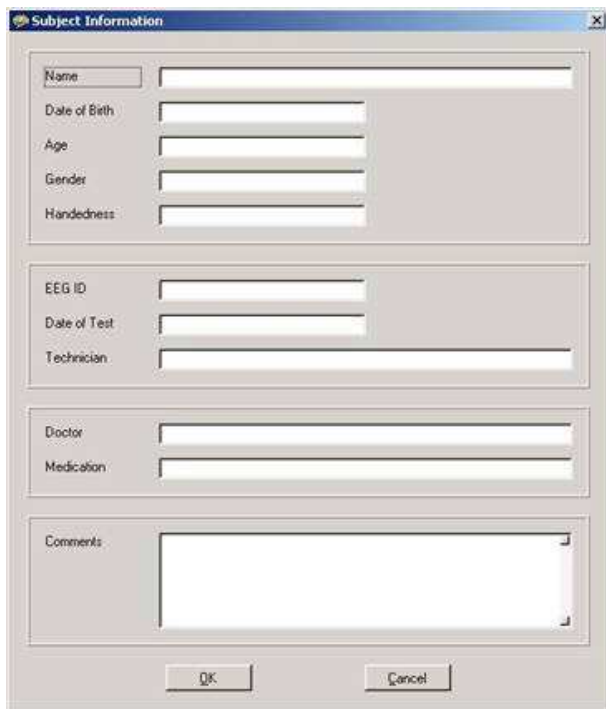
1a- Click File>Open>Lexicor>Lexicor NRS-24



1b Click No to the Lexicor edit import message



1c- Enter identifying information and then Click O.K.



The image shows a 'Subject Information' dialog box with the following fields:

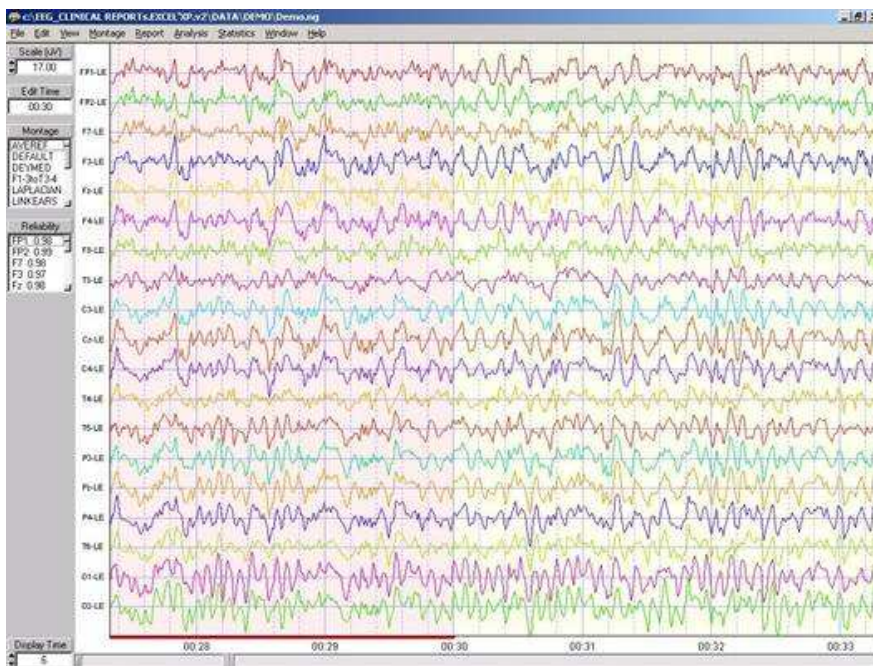
- Name: [Text Input]
- Date of Birth: [Date Picker]
- Age: [Text Input]
- Gender: [Text Input]
- Handedness: [Text Input]
- EEG ID: [Text Input]
- Date of Test: [Date Picker]
- Technician: [Text Input]
- Doctor: [Text Input]
- Medication: [Text Input]
- Comments: [Text Area]

Buttons: [OK] [Cancel]

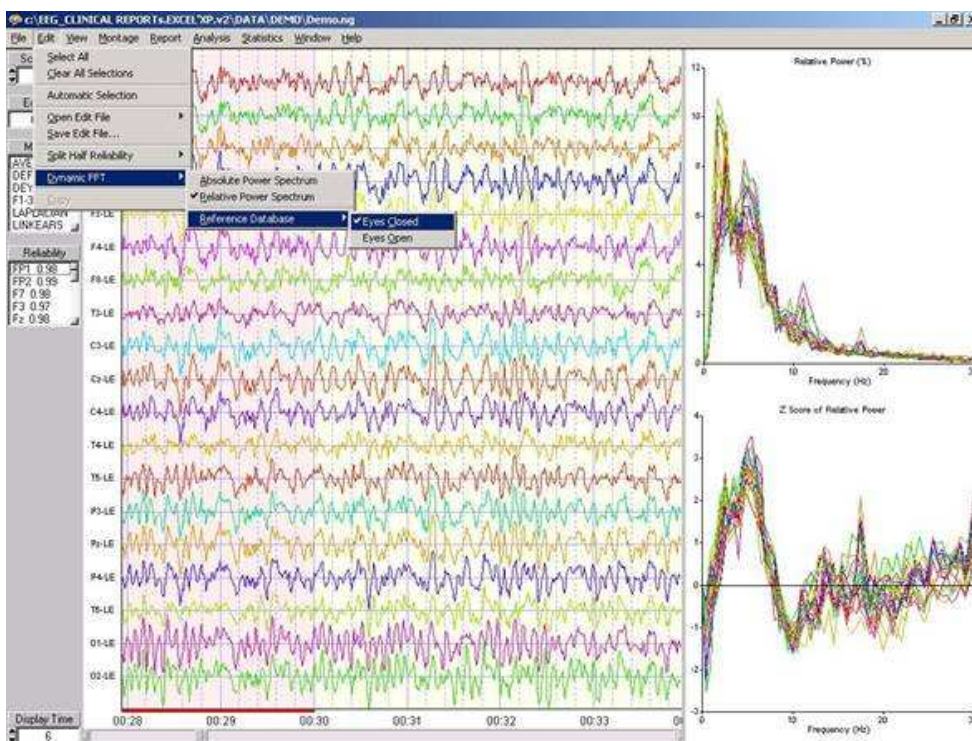
Step #2: Create NeuroGuide Analysis File. (For Demonstration Purposes Only)

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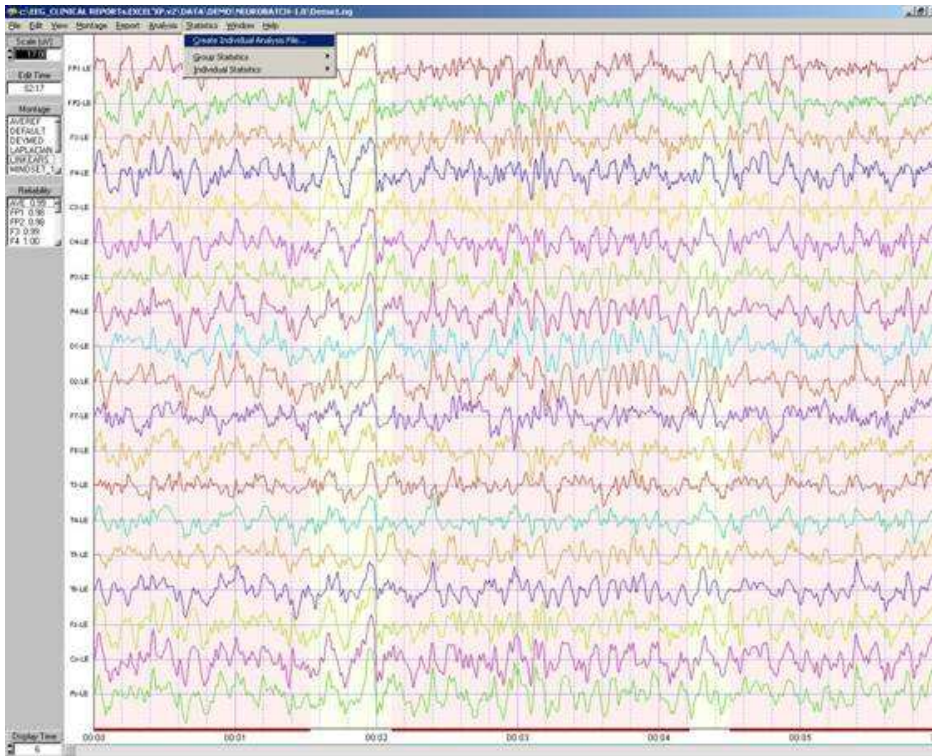
- 2a **Position the mouse and depress the left mouse button on the EEG tracings at time = 0 and slide to the right to select the first 30 seconds of EEG** (artifact is included and a blind selection of EEG is NOT recommended in practice).



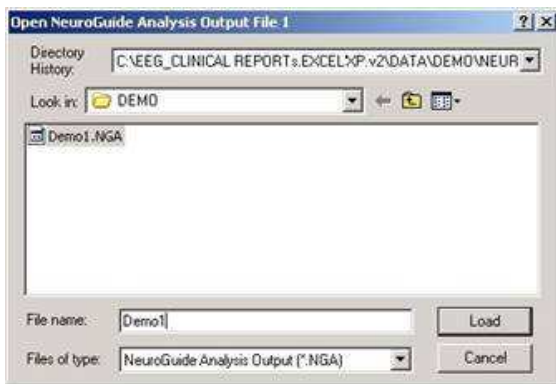
2b Click Edit>Dynamic FFT> check Relative Power to view the power spectra of your EEG edited selections.



2c - Click Statistics>Create NeuroGuide Analysis File:



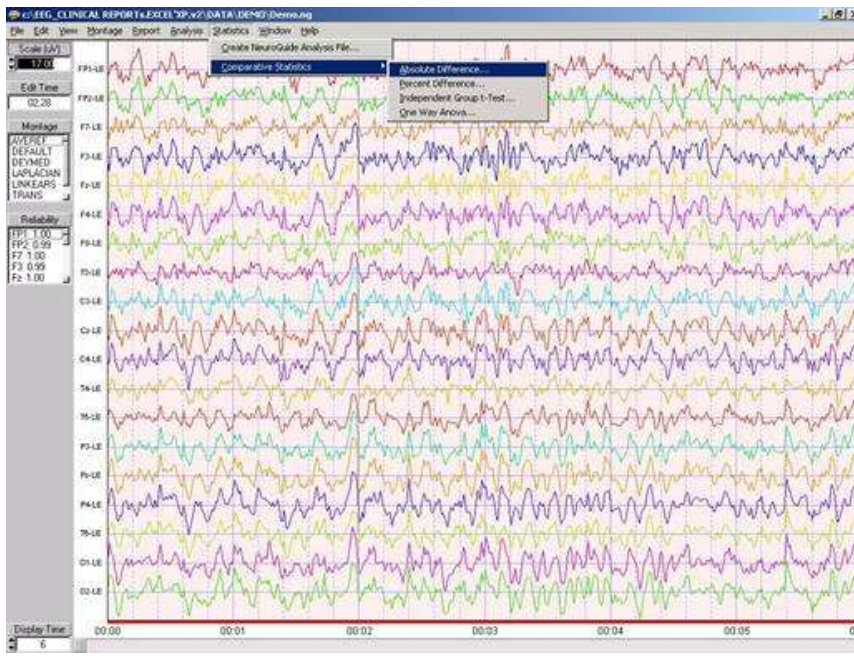
2d - Name the First Comparison NeuroGuide Analysis File: Demo1 and click Save.



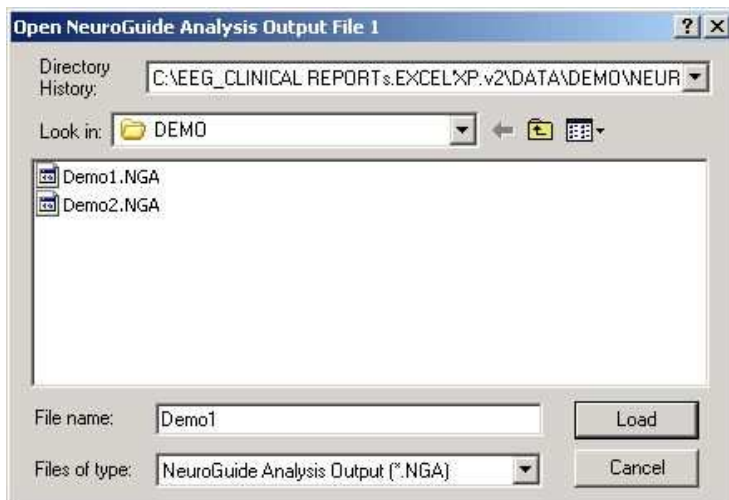
Step 3 - Compute Individual Statistics and Select Variables

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3a - Click Statistics>Individual Statistics>Absolute Difference



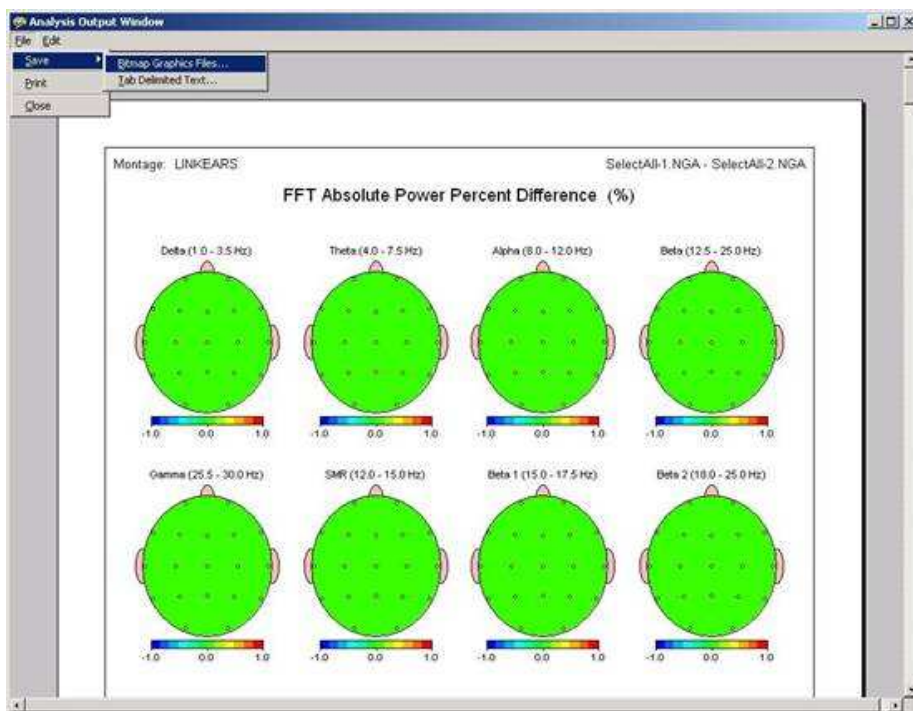
3b To test the statistics load the same file to compare it with itself. Load the first NeuroGuide Analysis File created in Step # 2d to be Compared: Demo1.NGA, when the window for File 2 appears then double click Demo1.NGA to compare the file with itself.



3c Select Analysis Options then Click O.K.:



3d- Topographic Difference Maps Green means Zero difference

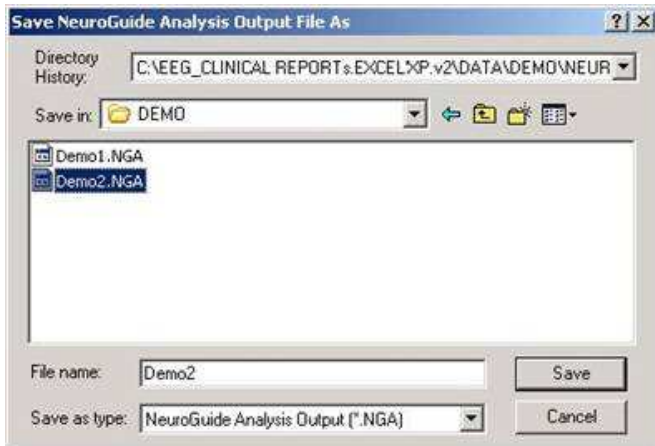


Step # 4 Repeat Step #3 by Comparing two different NeuroGuide Analysis Files. Close the Statistics output window. Then depress the left mouse button on the EEG tracings at 30 seconds and slide to add the successive 30 seconds of EEG by select the first 60 seconds of

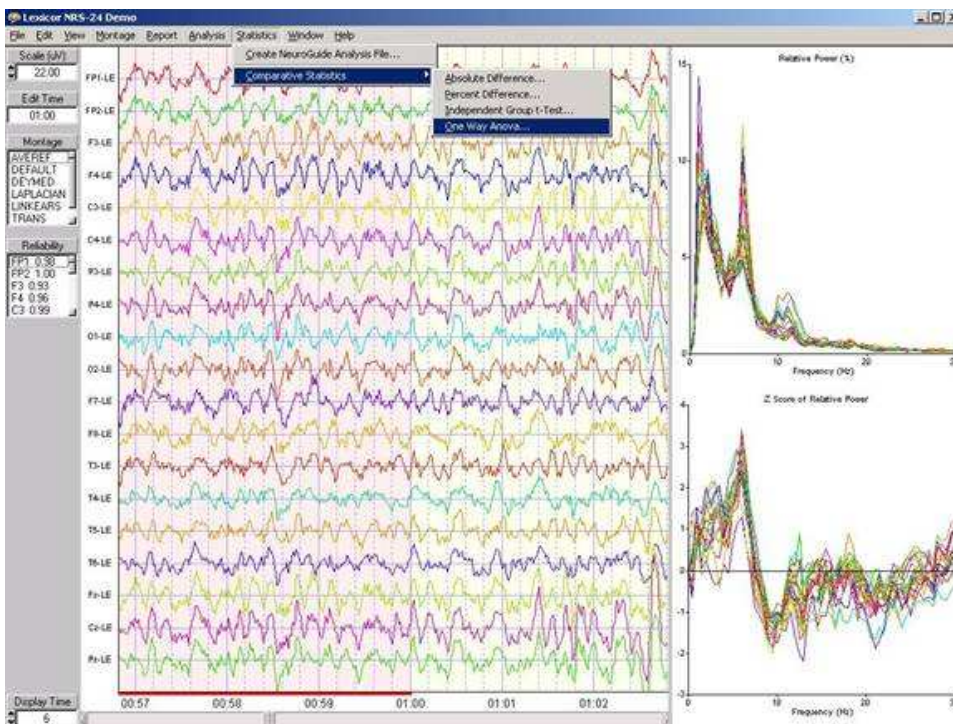
continuous time (0 to 1 minute).

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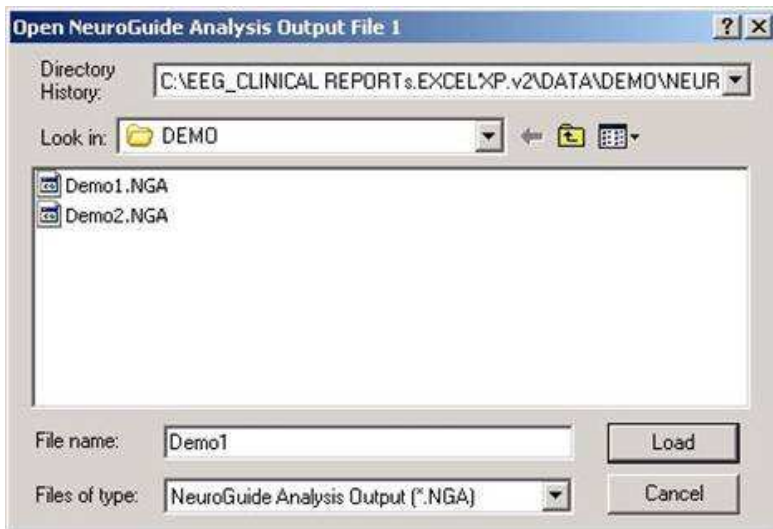
4a Click Statistics>Create NeuroGuide Analysis File and name the 60 second duration NeuroGuide Analysis File: Demo2



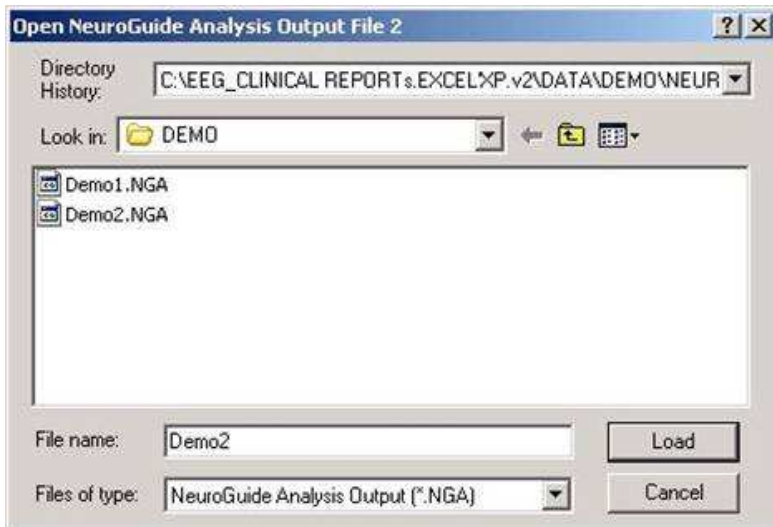
4b Click Statistics>Comparative Statistics>One Way ANOVA



4c Double click the First NeuroGuide Analysis File Demo1



4d Double click the second NeuroGuide Analysis File Demo2

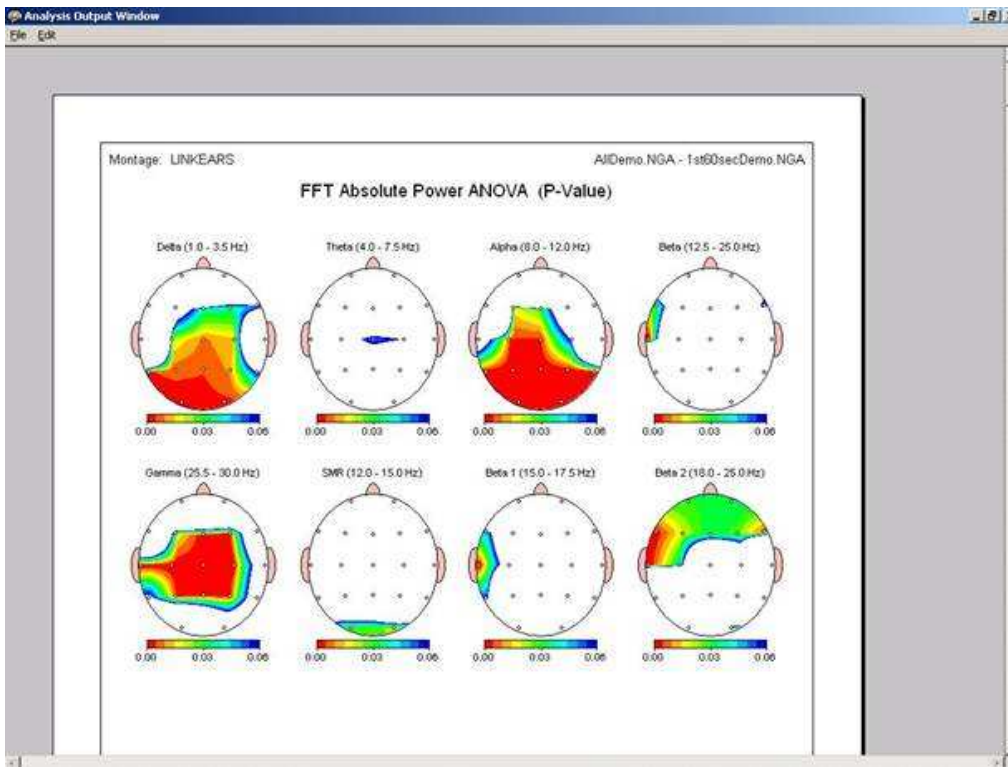


4e Click O.K. to the Analysis Options Window and NeuroGuide will begin to compute the statistics selected in step # 4b

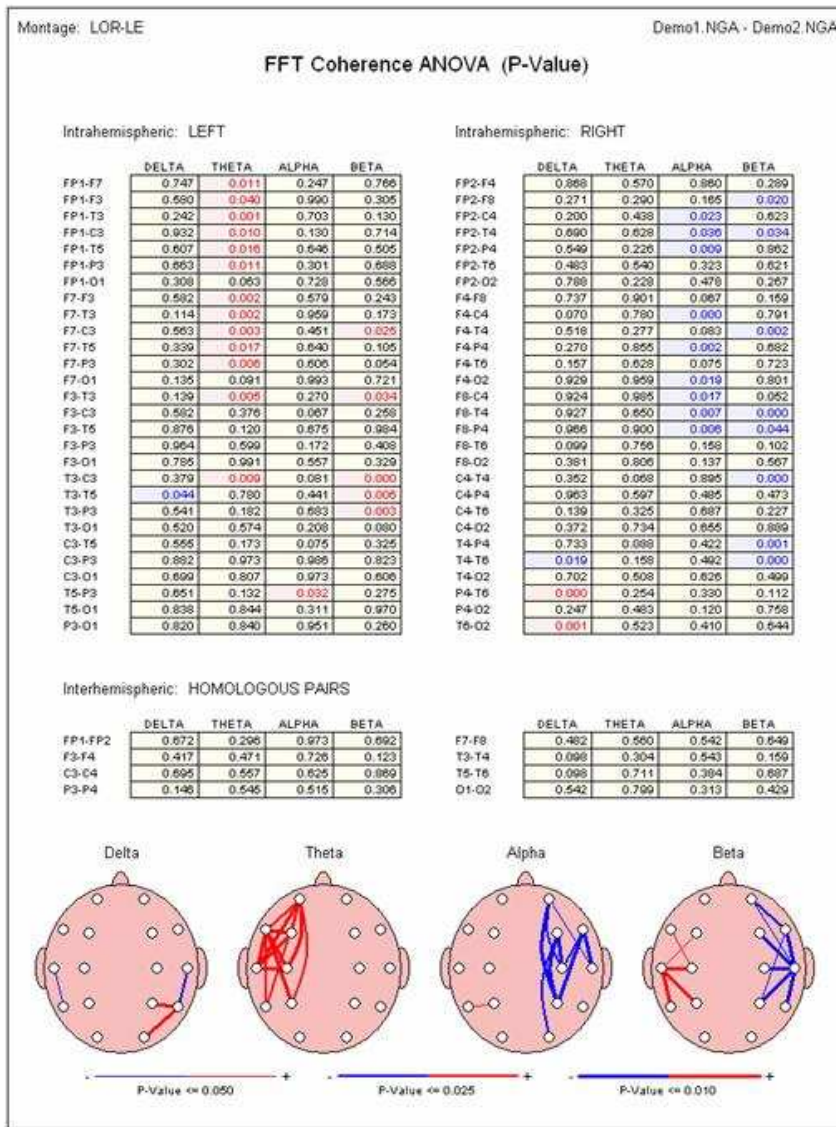


4f When NeuroGuide completes data processing an Analysis Output Window of P-Value of Differences of FFT Absolute Power Bands will be displayed. White = no statistically significant difference at $P < .06$, Red = $P < .001$ and $P < .05$ is Blue. This is a two-tailed P value and the direction of differences between Demo 1 and Demo2 is seen by examining Absolute Differences. Blue is the fringe statistic between $P < .05$ and $P < .06$.

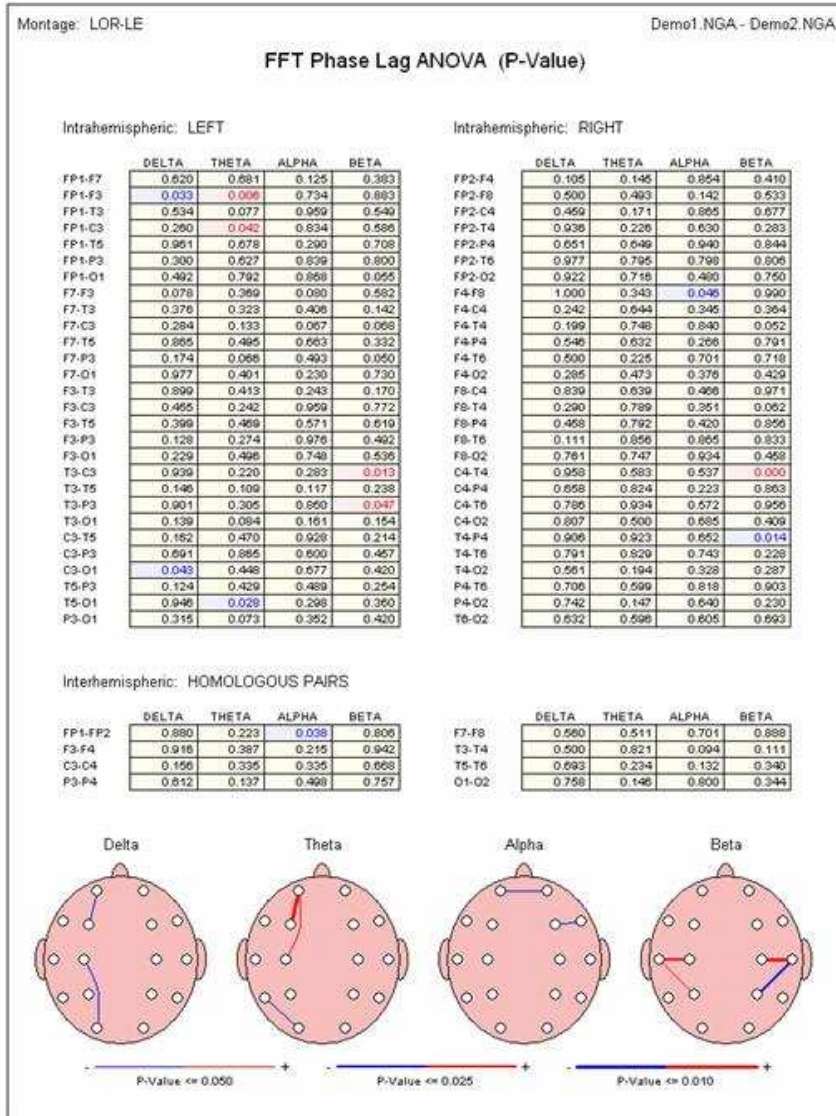
Move the down arrow key or the page down key or click the right margin wiper to view other comparative analyses.



4g - Example of Coherence P- Values in the ANOVA of Demo 1 and Demo 2



4h Example of EEG Phase P- Values in the ANOVA of Demo 1 vs Demo 2



4i Repeat Step #4 by selecting different statistics, e.g., Absolute Difference, Percent Difference and t-tests. Save and examine and experiment with different EEG selections in order to evaluate the sensitivity of different EEG selections. Save the Bitmaps and Text Data as in Step # 5.

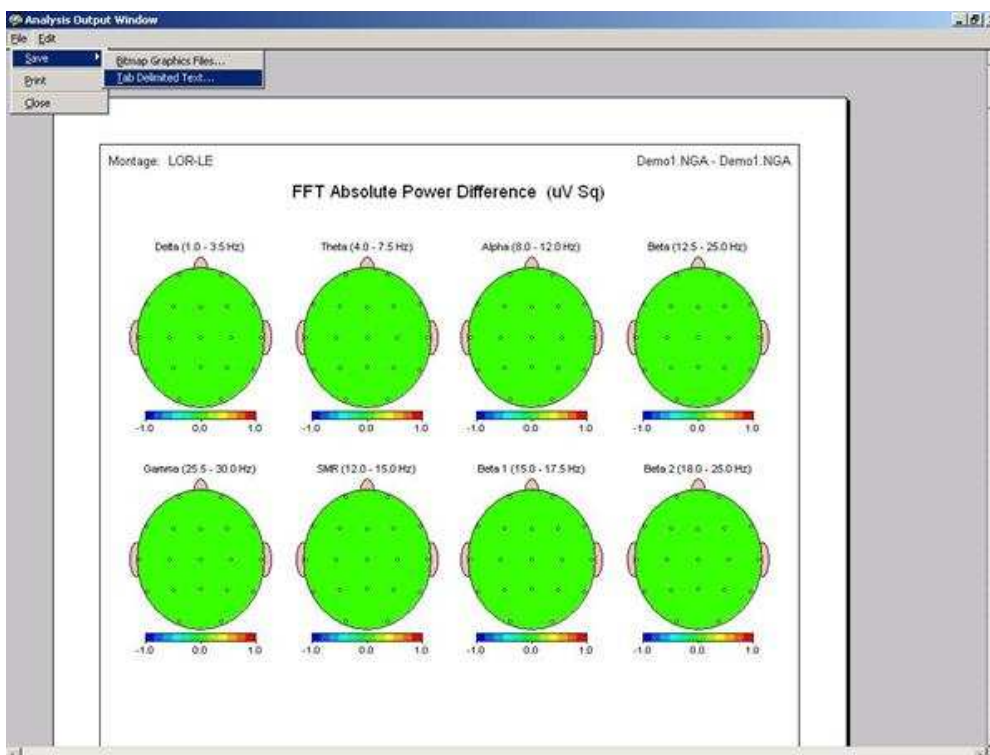
STEP # 5 Save Bitmaps and Text

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5a Save Bitmap Images, Click File Save>Bitmaps and create a folder e.g., bdemo1-LE (e.g., bitmap demo1 linked ears), open this folder, name the files, e.g., LEmaps and then click save:



5b - Save Text or ASCII Data,. Click File>Save>Tab Delimited Text (e.g., ascii demo1 linked ears) name file and then click save.



Step # 6 - Compute Group Statistics After Creating NeuroGuide Group

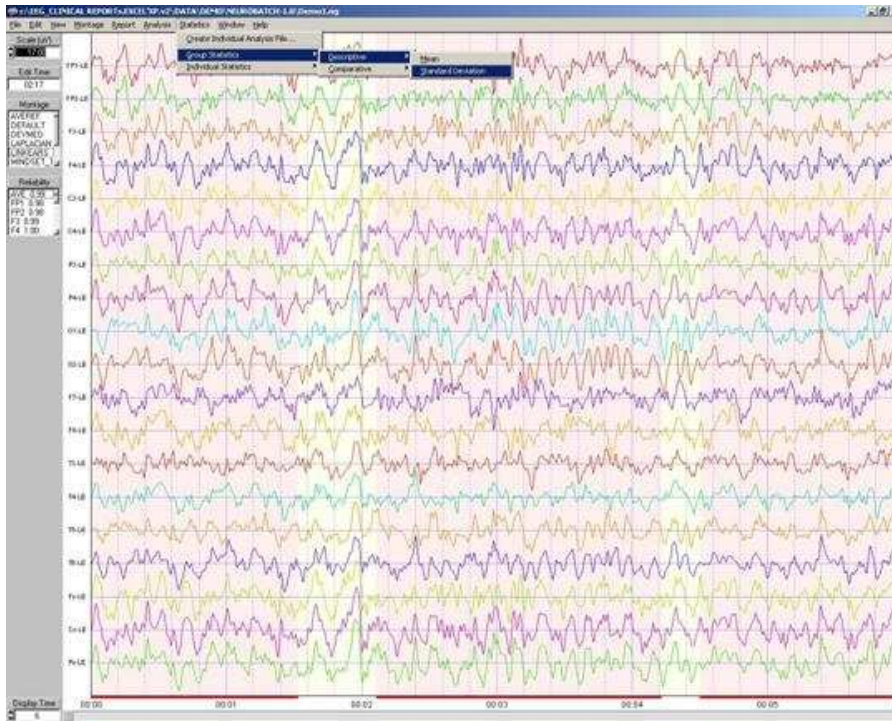
Analysis Files (*.NGG) using NeuroBatch

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6a Click Analysis>Batch and read the NeuroBatch manual which gives the step by step procedure to create Group Analysis Files *.NGG. NGG files are the input to the Group Statistics component of NeuroStat where NeuroBatch is used to create the group means and standard deviation values, etc. which are then utilized in NeuroStat.

NeuroStat imports the *.NGG files created by NeuroBatch and then computes the group statistics and color topographic displays and tab delimited outputs. NeuroBatch is required for NeuroStat group statistics only. Individual comparative statistics such as pre-treatment vs. post-treatment comparisons in Steps 4 and 5 do not require NeuroBatch.

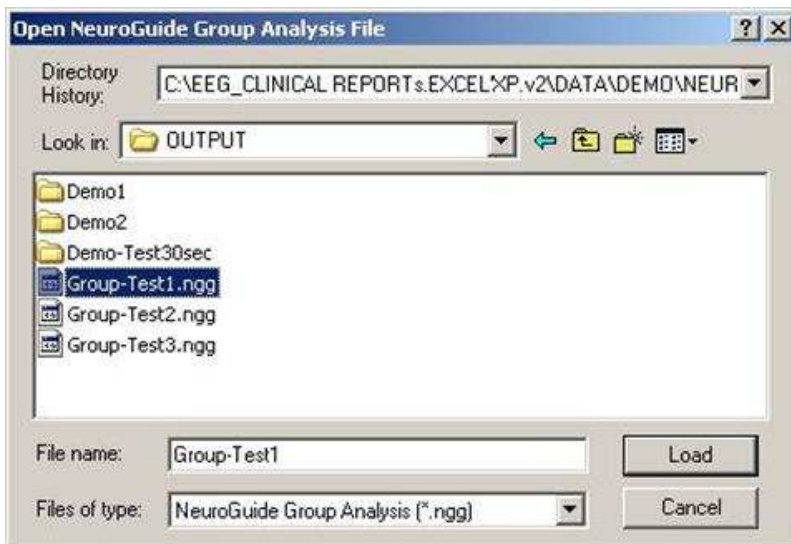
6b Click Statistics>Group Statistics>Descriptive>Standard Deviation



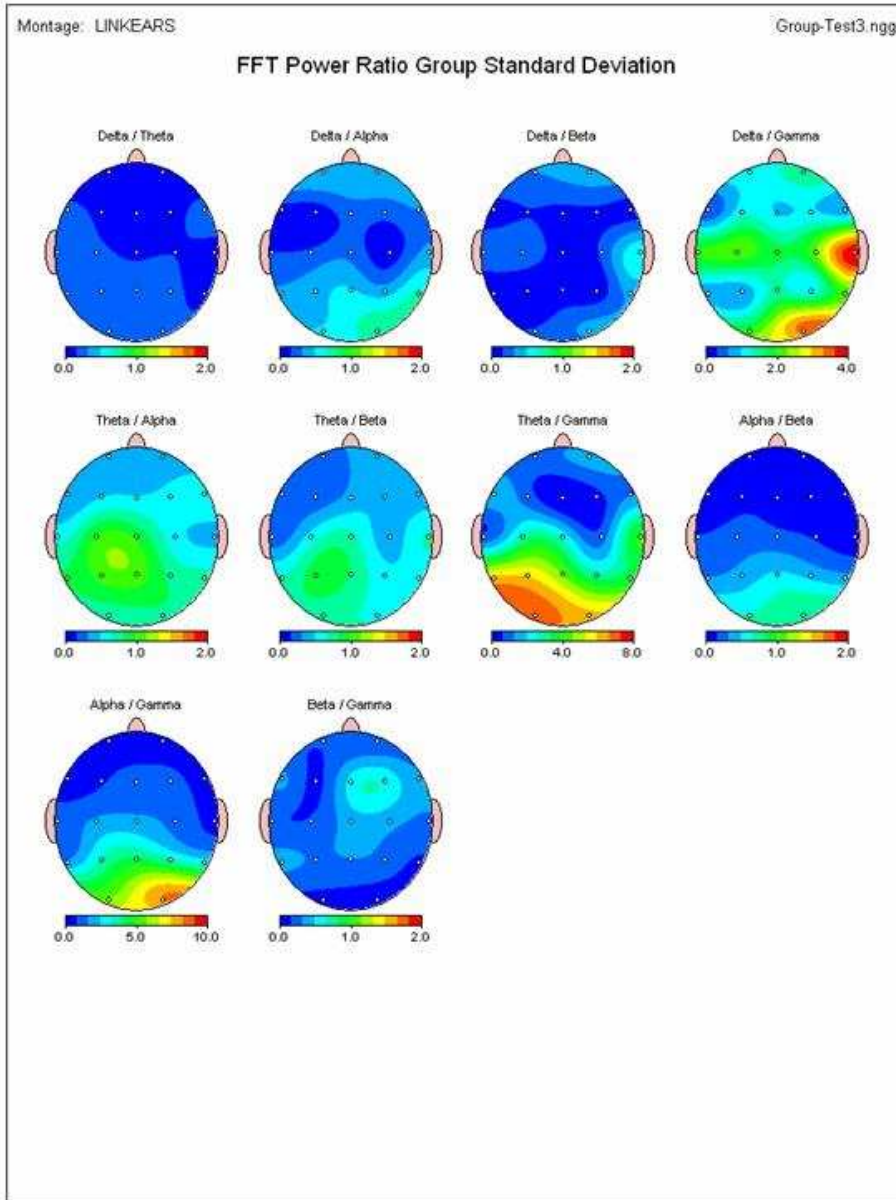
6c Click OK to the Analysis Option Window



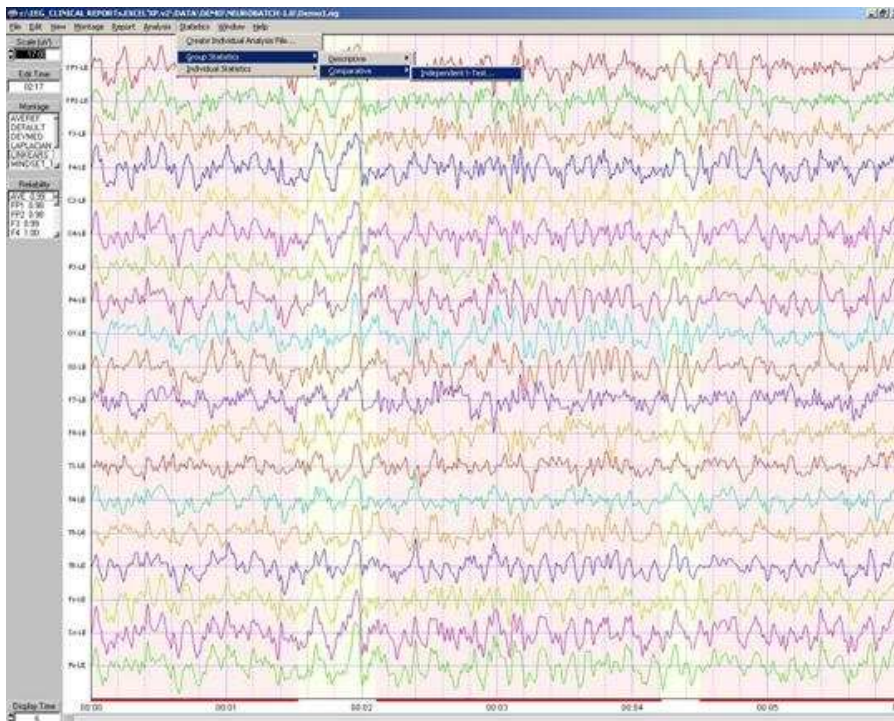
6d Double Click the Demo Group-1 NGG file and NeuroGuide will compute topographic maps and numerical tab delimited outputs of the Standard deviations.



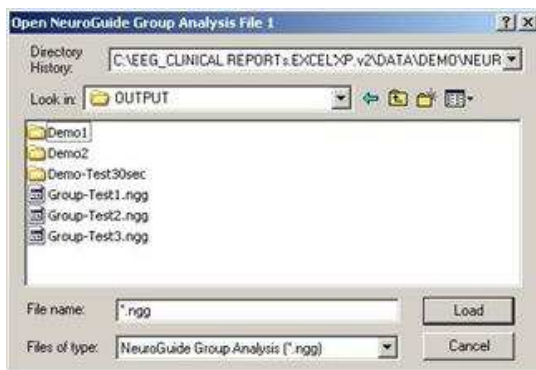
6e Example of Standard Deviation Color Topographic Maps. The values are the EEG values = 1 Standard Deviation.



6f To Compute Comparative Group Statistics Click Statistics>Group Statistics>Comparative> Independent t-Tests

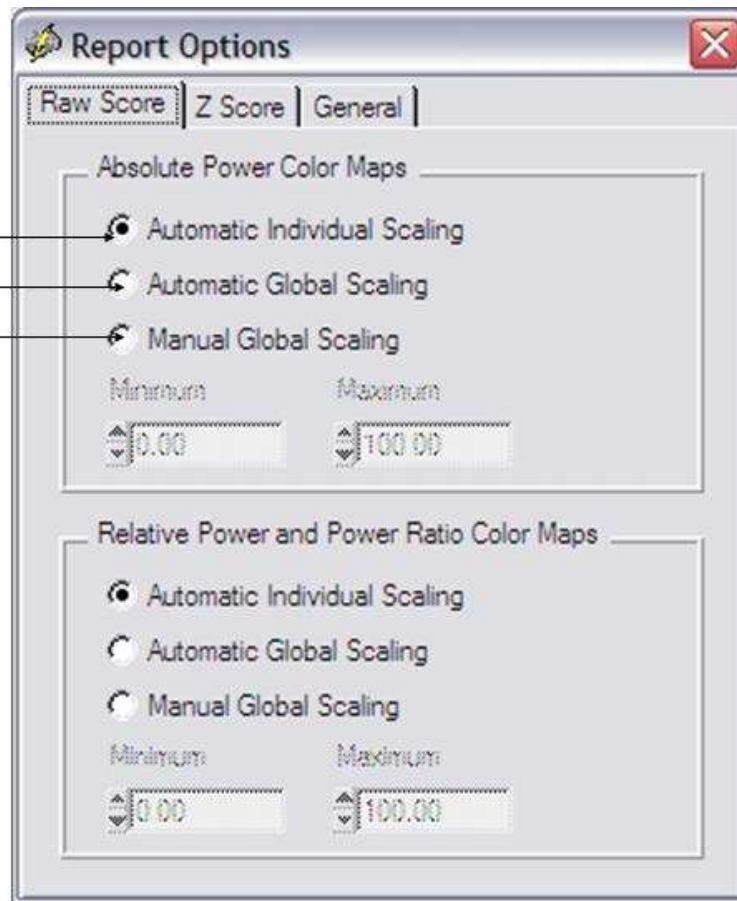


6g - After clicking OK to the analysis option window Double Click on the 1st group *.NGG file and then double click on the second *.NGG file in order to compute t-tests between the two groups



6h - Adjust the Scales of the Color Topographic Maps by clicking Report > Report Options and then selecting Automatic Individual Scaling where the minimum and maximum range is set for each individual map or Automatic Global Scaling where the min and max are computed for all maps or Manual Scaling in which the user sets the min and max.

Scale within individual Maps
Scale Maps Globally
Scale Maps Manually

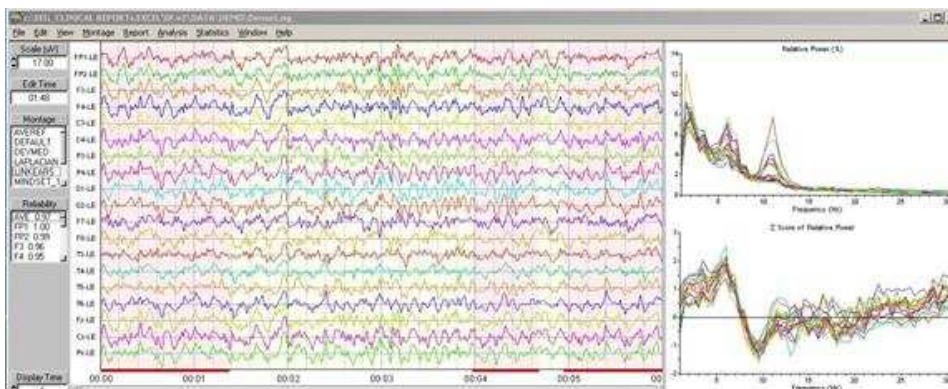


Step # 7 Simultaneously Launch Two NeuroGuides and Compare

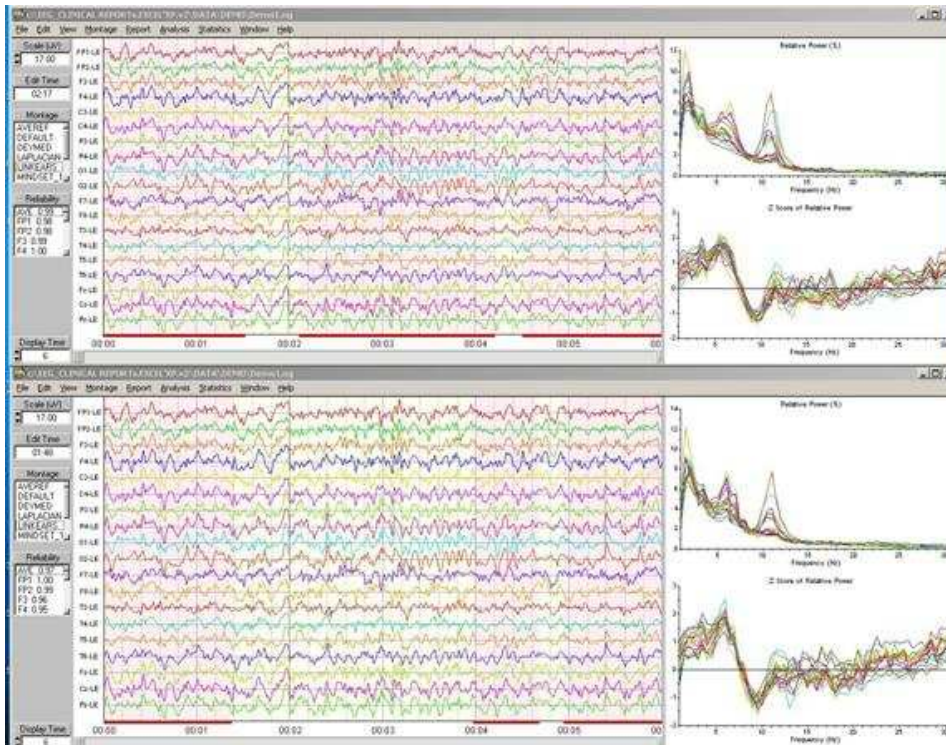
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This procedure is valuable because a single mouse click can toggle between two different EEG records and guide the creation of the NeuroGuide Analysis files *.NGA that are of interest for comparison purposes.

7a In the Demo mode Launch NeuroGuide as in Steps 1 to 2a. After selecting all, then resize the NeuroGuide edit window by clicking on the boundaries of the image and moving your mouse so as to position Test No. 1 in the top half of the computer screen



7b - Launch a Second NeuroGuide and repeat Steps 1 to 2 and Select All and resize the second NeuroGuide to fit into the bottom or top half of the computer screen.

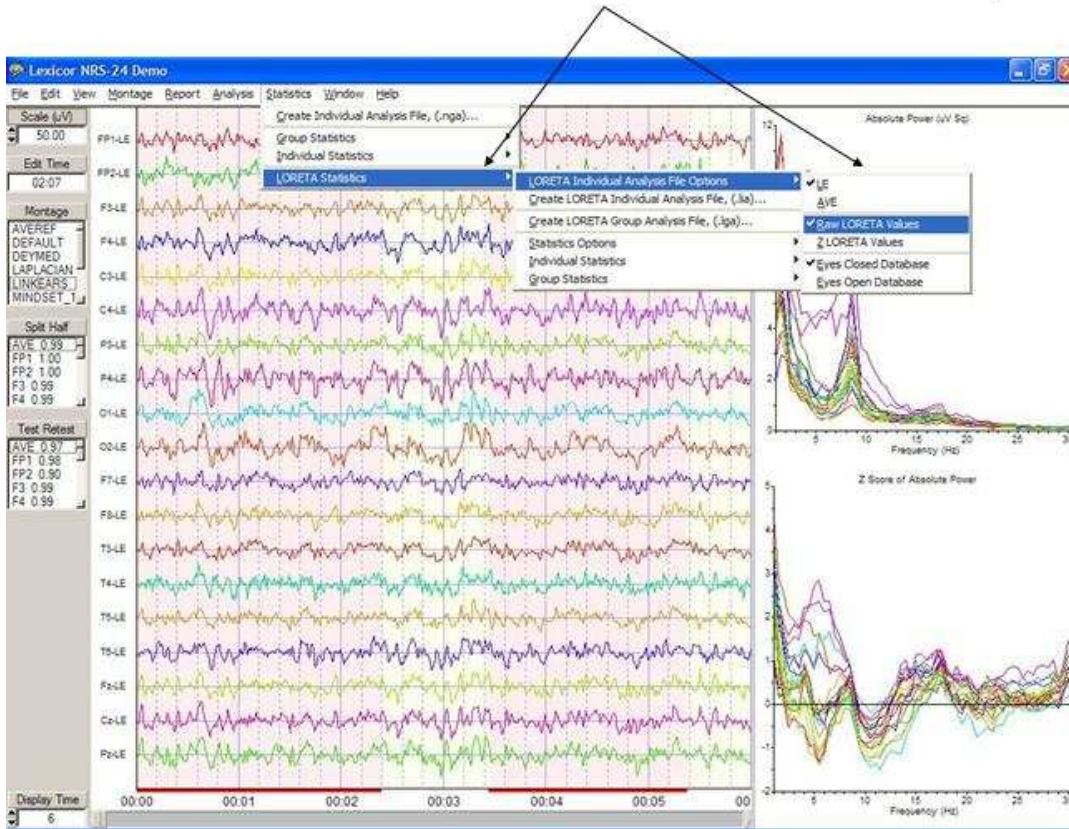


7c Toggle Between the Top and Bottom Window and Repeat Step # 2 to create different NeuroGuide Analysis Files *.NGA and then select a individual comparative test.

II- 3Dimensional LORETA: Individual Statistics

a- Launch NeuroGuide and open the Demo Lexicor NRS-24 file. Select the first 0 to 30 seconds and then click Statistics > LORETA Statistics > Create LORETA Individual Analysis File (*.lia). Name the file as 0-30sec.lia and then save.

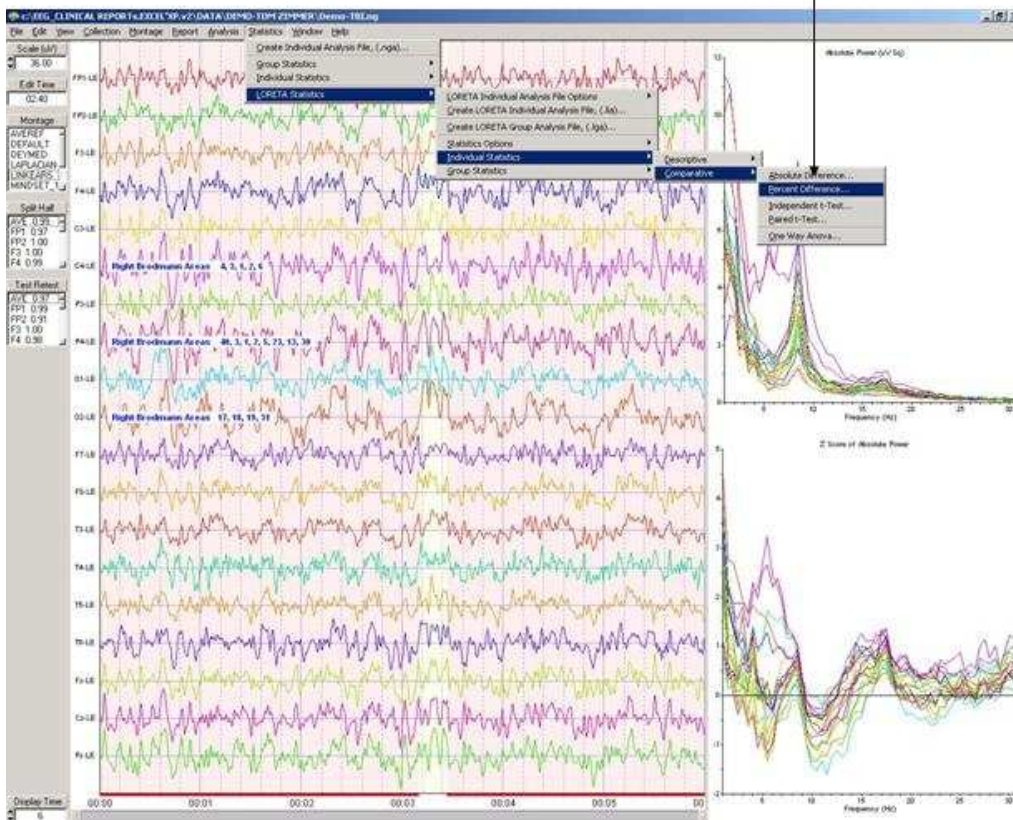
Click Statistics > LORETA Statistics > LORETA File Options



8b - Click Edit > Clear Selections and then depress the left mouse button and select EEG starting at 10 seconds and continuous to 40 seconds. Then click Statistics > LORETA Statistics > Create LORETA Individual Analysis File (*.lia). Name the file as 10-40sec.lia and then save.

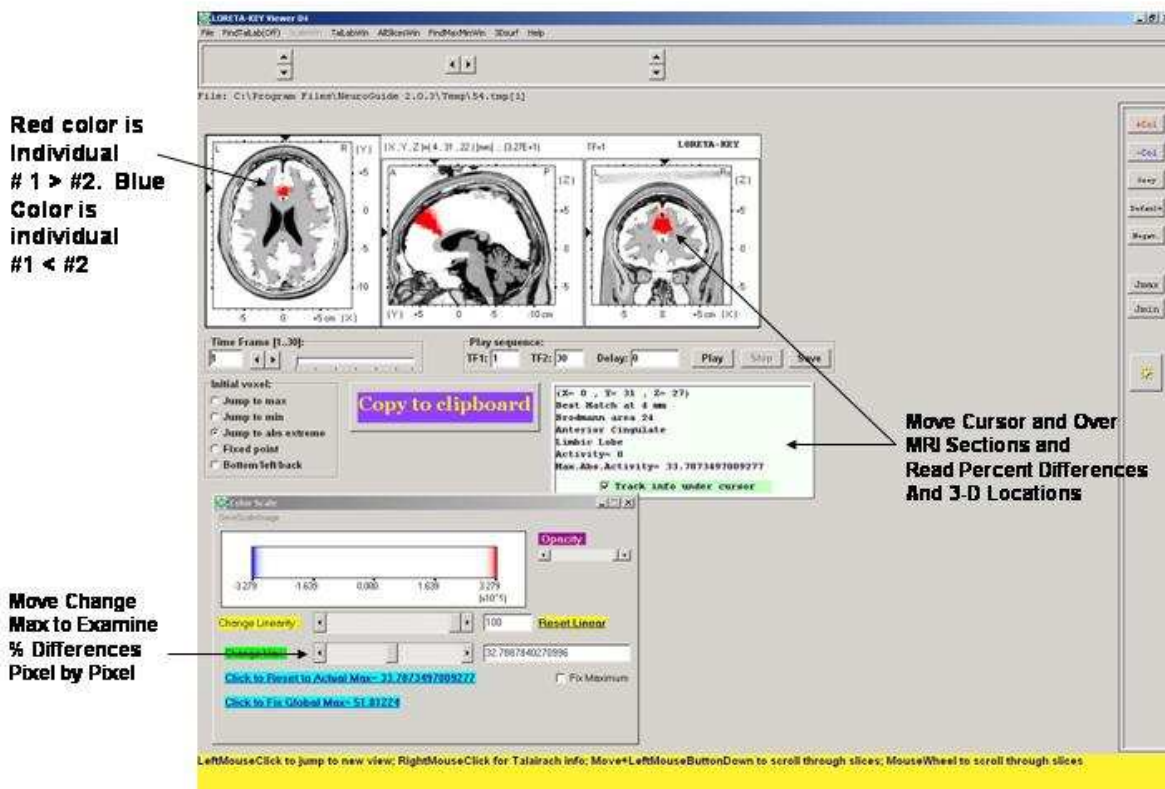
8c - Click Statistics > LORETA Statistics > Individual Statistics > Comparative > Percent Difference. Double click 0-30sec.lia in the Open LORETA Individual Analysis File 1 window. Then Double click 10-40sec.lia in the Open LORETA Individual Analysis File 2 window.

LORETA Individual Statistics > Comparative > Percent Differences



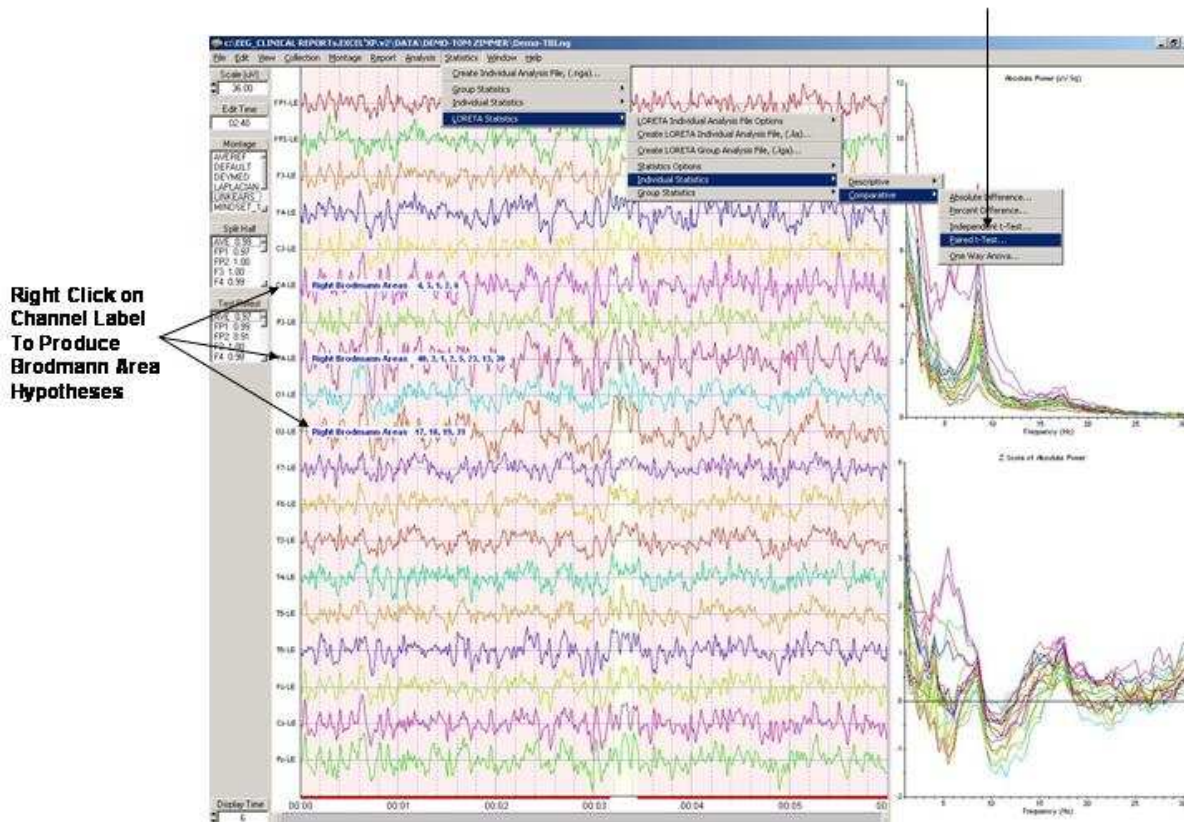
- 8d - A Save LORETA Statistics window will open so that one can save the results of the analysis in an ASCII *.lor format for purposes of importing into Excel or for use with the Key Institute software, etc. For purposes of this tutorial click Cancel to proceed to the computation of the statistic.**
- 8e - After the Key Institute software is launched, click ScaleWin on the Key Institute menu bar and position the color scale window in the lower left corner of the screen. Then advance through the frequencies by clicking the Time Frame right arrow to examine the percent differences between the 0 to 30 seconds of EEG versus the period from 10 seconds to 40 seconds of the EEG. Note that the maximum percent difference varies from about 30% at 1 Hz to about 9 % at 9 Hz.**

LORETA Key Institute Viewer of Percent Differences Between Two Individuals



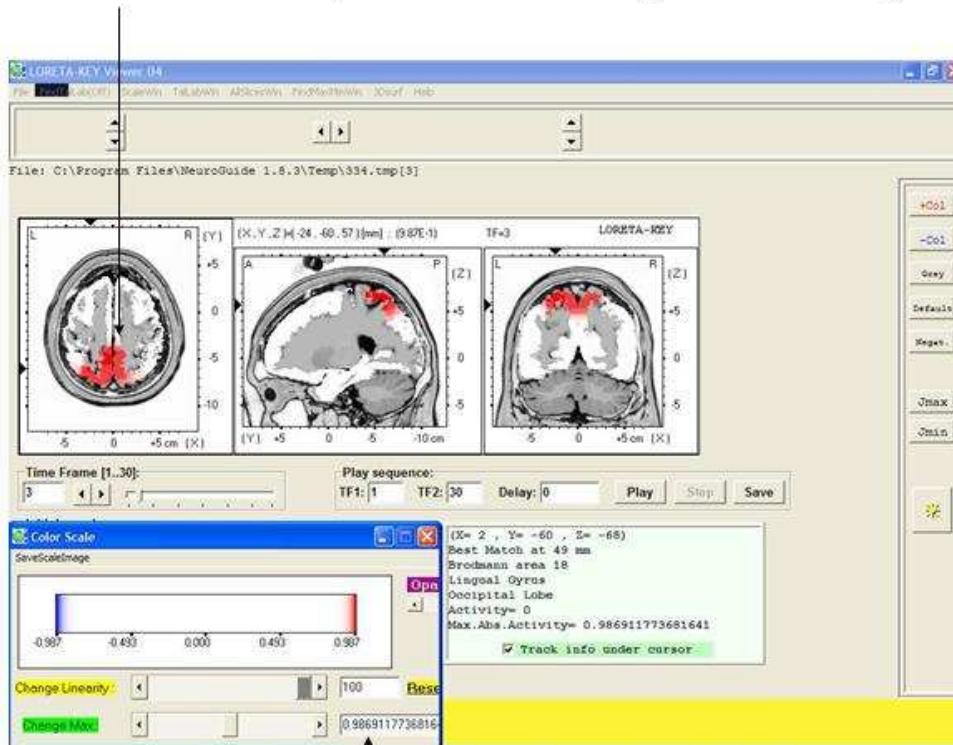
8f - Now click Statistics > LORETA Statistics > Individual Statistics > Comparative > Paired t-test. Double click 0-30sec.lia in the Open LORETA Individual Analysis File 1 window. Then Double click 10-400sec.lia in the Open LORETA Individual Analysis File 2 window. Click cancel when the Save LORETA Statistics window opens.

LORETA Individual Statistics Comparative Statistics Independent & Paired T-Tests



8g - After the Key Institute software is launched, click ScaleWin on the Key Institute menu bar and position the color scale window in the lower left corner of the screen. Move the Change Linearity wiper to the extreme right position or 100. For the moment keep the Change Max wiper in the default middle position so that the maximal probability values will be displayed. To evaluate statistical significance move the change max wiper to 1.0 and check the Fix Maximum box. Note that the Key Institute LORETA Viewer uses interpolation to color the MRI pixels which results in values greater than the maximum probability value, i.e., 1.0. Move the mouse over the regions of interest and read the exact probability values in the window at the bottom right of the viewer. Then click on the LORETA viewer that was launched in step 8e and compare the t-test probability value with the regions that had the highest percent differences in the first Viewer (step 8e). Advance through the frequencies by clicking the Time Frame right arrow to compare the two different statistics. Repeat step 8f by selecting a different statistic, such as independent t-tests or ANOVA. Also, evaluate the descriptive statistic option by clicking Statistics > LORETA Statistics > Individual Statistics > Descriptive to evaluate single *.lia files.

Examine P Values using a Paired T-Test – Eyes Closed vs Eyes Open



Move Change Max wiper to Examine Two Tail P Values

III - 3Dimensional LORETA: Group Statistics

- 9a - Launch NeuroGuide and open the Demo Lexicor NRS-24 file. Select the first 0 to 30 seconds and then click Statistics > LORETA Statistics > Create LORETA Individual Analysis File (*.lia). Name the file as Beginn1.lia and then save.
- 9b - Click Edit > Clear Selections and then depress the left mouse button and select EEG starting at 10 seconds and continuous to 40 seconds. Then click Statistics > LORETA Statistics > Create LORETA Individual Analysis File (*.lia). Name the file as Beginn2.lia and then save.
- 9c - Click Edit > Clear All Selections and then click the End Key. Position the mouse on the last EEG values and depress the left mouse button and select EEG starting at 3.48 seconds and move the mouse to the right and stop at 3:18 seconds. Then click Statistics > LORETA Statistics > Create LORETA Individual Analysis File (*.lia). Name the file as End1.lia and then save.
- 9d - Click Statistics > LORETA Statistics > Create LORETA Group Analysis File (*.lga). In the LORETA Group Analysis File Creation window Click browse to browse to the folder where the individual analysis files were saved (i.e., the *.lia files) and click Done.
- 9e - In the LORETA Group Analysis File Creation window click the file Beginn1.lia and click Add. Then click the file Beginn2.lia and click Add. Then Click O.K. In

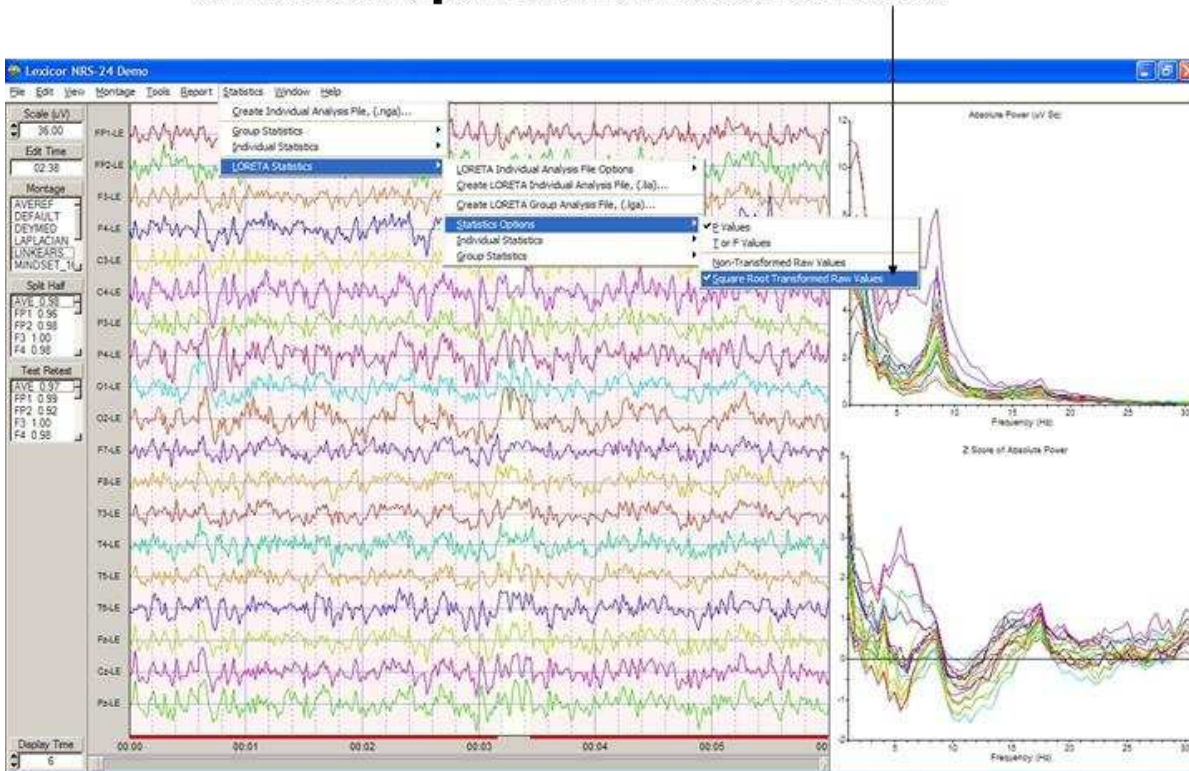
the Save LORETA Group Analysis Output File As name the Group file Beginning.lga.

9f - Now let us create the second group file. Click Statistics > LORETA Statistics > Create LORETA Group Analysis File (*.lga). In the LORETA Group Analysis File Creation window Click browse to browse to the folder where the individual analysis files were saved (i.e., the *.lia files) and click Done.

9g - In the LORETA Group Analysis File Creation window click the file End1.lia and click Add. Then click the file End2.lia and click Add. Then Click O.K. In the Save LORETA Group Analysis Individual Output File As name the Group file End.lga.

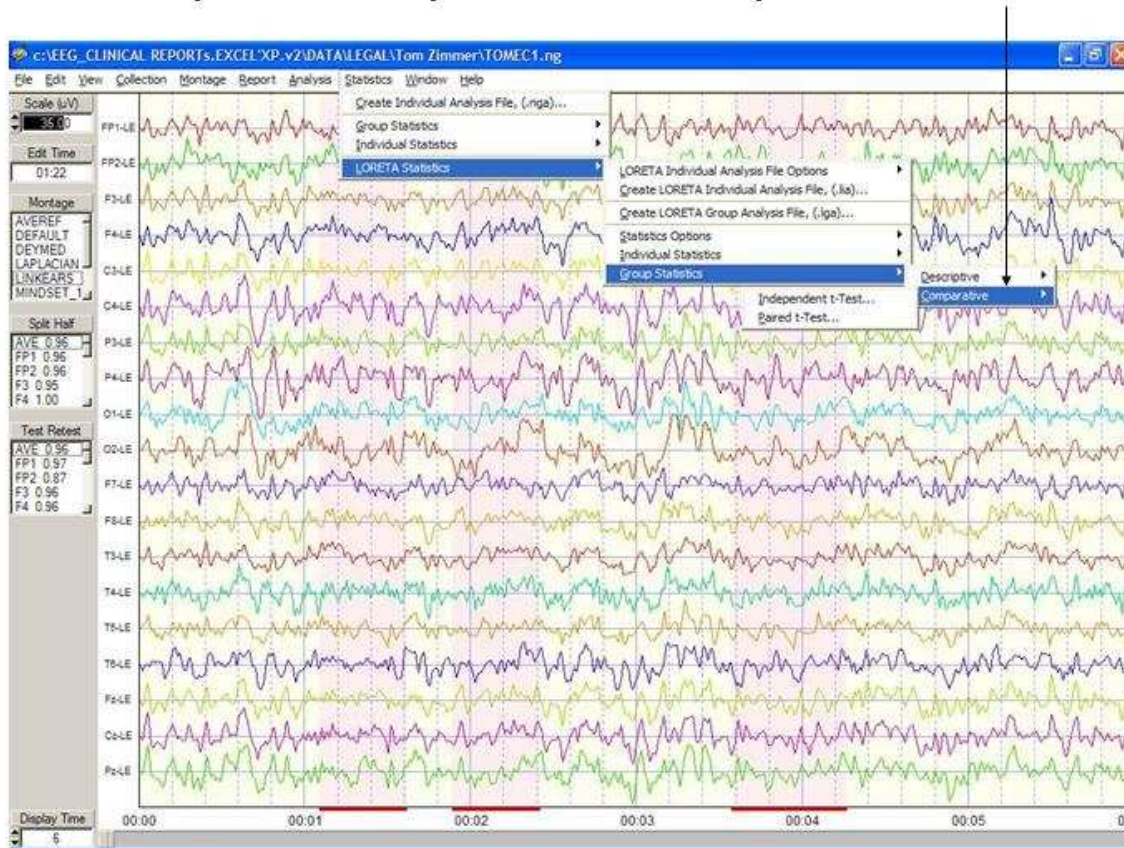
9h - Click Statistics > LORETA Statistics > Statistics Options in order to select P values or T values as well as the non-transformed raw values (squared source current vectors (square root of the sum of squares of the x, y & z moments) or the square root transformed raw values which will return source current values in units of amperes/meter squared.

Click Statistics > LORETA Statistics > Statistics Options > Non-Transformed Raw Values or Square Root Transformed Raw Values



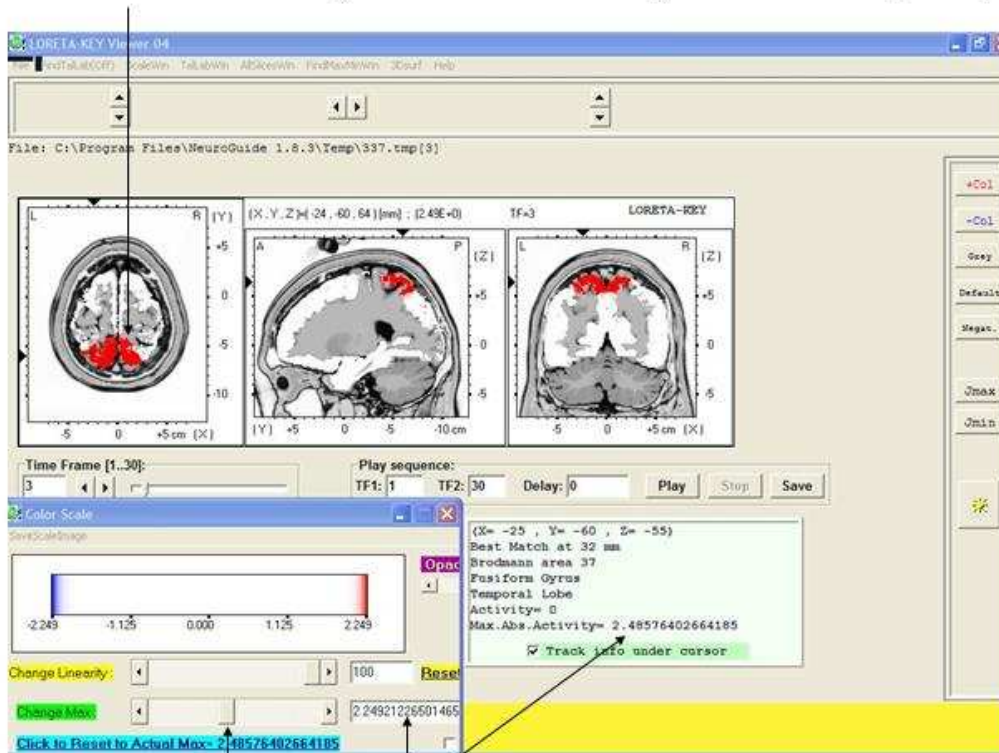
9i - Click Statistics > LORETA Statistics > Group Statistics > Comparative > Paired t-test. Select by double clicking the file Beginning.lga, then double click on the file End.lga. A Save LORETA Statistics window will open so that one can save the results of the analysis in an ASCII *.lor format for purposes of importing into Excel or for use with the Key Institute software, etc. For purposes of this tutorial click Cancel to proceed to the computation of the statistic.

LORETA Group Statistics Comparative Statistics Independent & Paired T-Tests



- 9i - After the Key Institute software is launched, click ScaleWin on the Key Institute menu bar and position the color scale window in the lower left corner of the screen. Move the Change Linearity wiper to the extreme right position or 100. For the moment keep the Change Max wiper in the default middle position so that the maximal probability values will be displayed. To evaluate statistical significance move the change max wiper to 1.0 and check the Fix Maximum box. Note that the Key Institute LORETA Viewer uses interpolation to color the MRI pixels which results in values greater than the maximum probability value, i.e., 1.0. Move the mouse over the regions of interest and read the exact probability values in the window at the bottom right of the viewer. Advance through the frequencies by clicking the Time Frame right arrow to compare the two different statistics. Repeat step 9h by selecting a different statistic, such as independent t-tests. Also, evaluate the descriptive statistic option by clicking Statistics > LORETA Statistics > Group Statistics > Descriptive to evaluate single *.lga files.

Examine T Values using a Paired T-Test – Eyes Closed vs Eyes Open



Move Change Max wiper to Examine Two Tail T Scores and not just the P values

Appendix A:

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Computation of the auto-spectral and cross-spectral densities of the edited EEG selections

- 1- The FFT parameters are: epoch = 2 seconds at a sample rate of 128 sample/sec = 256 digital time points and a frequency range from 0.5 to 40 Hz at a resolution of 0.5 Hz using a cosine taper window to minimize leakage. Each 2 second FFT is 81 rows (frequencies 0 to 40 Hz) X 19 columns (electrode locations) = 1,539 element cross-spectral matrix for each subject.
- 2- In order to minimize the effects of windowing in the FFT (Kaiser and Serman, J. Neurotherapy, 4(3): 85-92, 2001) a EEG sliding average of the 256 point FFT cross-spectral matrix was computed for each normal subjects edited EEG by advancing in 64 point steps (75% overlap) and recomputing the FFT and continuing with the 64 point sliding window of 256 point FFT cross-spectrum for the entire edited EEG record. Each of the 81 frequencies for each 19 channels is \log_{10} transformed to better approximate a normal distribution. The total number of 2 second windows is the number that is entered into the analysis of variance and t-tests and it is used to compute the degrees of freedom for a given statistical test.
- 3- A mean, variance, standard deviation, sum of squares, and squared sum of the real (cosine) and imaginary (sine) coefficients of the cross-spectral matrix is computed across the sliding average of edited EEG for all 19 leads for the total number of 81 and 1,539 log

transformed elements for each subject. This creates the following six basic spectral measurement sets and their derivatives 1- Cross-Spectral Power (square root of the sums of squares of the real and imaginary coefficients); 2- Auto-Spectral Power which is the diagonal of the cross-spectral matrix where the imaginary coefficient = 0 and power = sine square; 3- Coherence = square of the cross-spectrum divided by the product of the two auto-spectra; 4- Phase = arctangent of the ratio of the real/imaginary components for frequencies from 0.5 to 40 Hz;; 5- Real coefficients; 6 Imaginary coefficients.

4- The results of the computations are stored in the NeuroGuide Analysis File, designated as *.NGA. These results are used in the comparative statistical analyses when one selects and opens the .NGA files in the menu operation of Statistics>Comparative Statistics. Calibration of NeuroStat is by computing the statistics contained in the NRS-24 demo sample of EEG from an unidentified traumatic brain injured patient.

5- The results of the LORETA computations from a single selection of EEG are stored in a LORETA individual analysis file *.LIA. The .lia file contains all of the cross-spectral values for each 2 second epoch so that the means and standard deviations, higher statistical moments and sum of squares, etc. can be computed for each gray matter pixel in the Montreal Neurological normative MRI. Details of the computation of the T matrix and the J currents as the square root of the sum of the squares (x, y & z) is described in Appendix F in the NeuroGuide manual. The mathematics for the LORETA statistics are identical to the surface EEG statistics except there is a larger number of individual comparisons (2,393) in comparison to the surface 19 channel situation.

6- This issue of multiple comparisons when using LORETA is best handled by the statistics of "Planned Comparisons" or where one first creates hypotheses as to frequencies and Brodmann areas before launching LORETA. For example, if the surface EEG at F3 is > 2 standard deviations deviant from normal at 4 Hz, then the hypothesis prior to launching LORETA is significant deviations from normal in left Brodmann areas 8 & 9 at 4 Hz. Bonferroni corrections (i.e., alpha/n) are blind exploratory adjustments and result in large Type II errors. Hypothesis testing is preferred over multiple comparison adjustments.

Appendix B: Equations

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1- **Absolute Differences:** Mean test 1 Mean Test 2 for 19 channels and 81 frequencies or in the case of LORETA for 2,394 gray matter pixels from 1 to 30 Hz in 1 Hz increments (by averaging 0.5 Hz bands).

2- **Percent Differences:** $\frac{\bar{X}_{test1} - \bar{X}_{Test2}}{\bar{X}_{test1} + \bar{X}_{Test2}} \times 100$, where X bar = mean

3- **Independent t test (Assuming Population Variance Not Equal)** (from Winer, R.J., Statistical Principles in Experimental Design, McGraw-Hill, New York, 1962, p. 42 43):

$t = \frac{(\bar{X}_1 - \bar{X}_2) - (u_1 - u_2)}{\sqrt{(s_1^2/n_1) + (s_2^2/n_2)}}$, To test the hypothesis that $u_1 - u_2 = 0$ against a two-tailed alternative hypothesis $P < .025$.

- 4- Paired t-test (from Winer, R.J., Statistical Principles in Experimental Design, McGraw-Hill, New York, 1962, p. 44-46):

$$t = \frac{\bar{d} - (u_1 - u_2)}{\sqrt{s_d^2/n}}$$

, where \bar{d} = the mean differences between test 1 and test 2 and s_d^2 is the variance of the differences and n = number of FFT windows in test 1 + test 2.

- 5- One Way Analysis of Variance (from Hayes, W.J. Statistics for the Social Sciences, Holt, Rinehart and Winston, Inc., New York, 1973, p. 473-475):

$$\text{Sum of squares total (SS total)} = \sum_j \sum_i (y_{ij} - M)^2$$

$$\text{Sum of squares within (SS within)} = \sum_j \sum_i (y_{ij} - M_j)^2$$

$$\text{Sum of squares between (SS between)} = \sum_j \sum_i (y_{ij} - M_i)^2$$

Where M = mean and j = number of groups to be compared, i.e., $j = 2$ and i = number of FFT windows in both test 1 and test 2. Degrees of freedom = $J - 1$ and $i - 2$ and F = Mean square between groups/mean square within groups. Probability is computed using the F based on 1 and $i - 2$ degrees of freedom.

- 6 - Repeated Measures Analysis of Variance (from Hayes, W.J. Statistics for the Social Sciences, Holt, Rinehart and Winston, Inc., New York, 1973, p. 571-573):

$$\text{Sum of squares between (SS between)} = \frac{\sum_i (\sum_j y_{ij})^2}{J} - \frac{(\sum_i \sum_j y_{ij})^2}{nJ}$$

$$\text{Sum of squares within (SS within)} = \sum_i \sum_j y_{ij}^2 - \frac{\sum_i (\sum_j y_{ij})^2}{J}$$

$$\text{Sum of squares between treatments} = \frac{\sum_i (\sum_j y_{ij})^2}{n} - \frac{(\sum_i \sum_j y_{ij})^2}{nJ}$$

SS residual SS within SS between treatments. J = number of groups (2) and n =

number of FFT windows in both test 1 and test 2. The degrees of freedom are 1 and n - 2.

7- Covariance, Correlation and R^2 (from Winer, R.J., Statistical Principles in Experimental Design, McGraw-Hill, New York, 1962, p. 44 52

):

$$L_1 = n \sum X_1^2 - (\sum X_1)^2 \quad \text{and} \quad L_2 = n \sum X_2^2 - (\sum X_2)^2$$

$$L_{12} = n \sum X_1 X_2 - (\sum X_1)(\sum X_2) = \text{Covariance}$$

$$r_{12} = \frac{L_{12}}{\sqrt{L_1 L_2}} = \text{Correlation}$$

$$r_{12}^2 = R^2 \text{ or the percent variance shared by test 1 and test 2}$$

All comments and feedback are welcome.

Contact us at qeeg@appliedneuroscience.com and tell us what you think.