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Improving the methodology for non-invasive autonomic nervous system recording and its implementation in behavioral research

van Lien, R.

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E-mail address:

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APPENDIX III

Example of a VU-AMS workshop with assignments

Program overview

Introduction to recording with the VU-AMS and analyzing with DAMS.

Today I am wearing the VU-AMS to provide a fresh recording that we can use to get familiar with the basics while explaining some new features of DAMS 3.0. We will process this file together in a plenary session before we continue with some hands on scoring in the individual sessions. New DAMS features in the plenary session are:

1. VU-AMS 5 lead version
2. Bluetooth connection for online wireless heart rate monitoring
3. Instant Heart Rate during online mode
4. Remote event marker via Bluetooth
5. Automated Scoring for ECG QRST Points
6. Option to remove all HIGH & MEDIUM Suspicious Beats in Detect R-Peaks Tab

Focus on the PEP

After the introduction to DAMS 3.0 we are going to inspect/correct the impedance and respiration scoring of several pre-labeled recordings. The files used in this part of the workshop are from an ambulatory experiment where subjects were guided through common posture and activity changes accompanied by an experimenter. We will come across some new impedance scoring features while doing this.

All participants will score the same recordings and compare them for inter-rater reliability. We will merge data in pairs to see what the consensus between the scoring of two raters was. We will discuss the deviant cases.

24H recordings

We continue with PEP scoring and respiration and RSA scoring on ambulatory data collected in a naturalistic setting. This kind of data usually consists of more and longer complexes and is often prone to noise which makes scoring of the impedance signal quite a challenge!

SCL analyses now available!

For the first time in DAMS we have the opportunity to experience the new SCL analyses consisting of Label and stimulus based export of Skin Conductance Data. We treat this subject separate as it has a different output from ECG and ICG analyses.

Wrap up: some additional features worth mentioning and time for Questions.

At this point we will have seen a lot of data and discussed quite a lot but there are some features worth mentioning.

- Divide a particular label into time labels of chosen interval.
- Batch Export of Raw Signals with & without label information
- Batch Analyze & Export Data (ASCII & Excel) for automatic scoring of ECG & ICG Complexes

Working with DAMS

Step 1: Opening the AMS recording for the first time (.5fs file).

Upon opening the 5fs file, the Inspect Data tab simply gives you an overview of your data. Above the recording time, the Inter Beat Interval (IBI) time series is given as extracted from the ECG. The clock time and IBI signal of the entire recording are presented in all analysis tabs, and functions as orientation point. Vertical lines represent the times at which the event button was pressed.

Assignment 1. Open the example file with the DAMS 3.0 program.

**The example file is named Morning_Recording_Rene.5fs.*

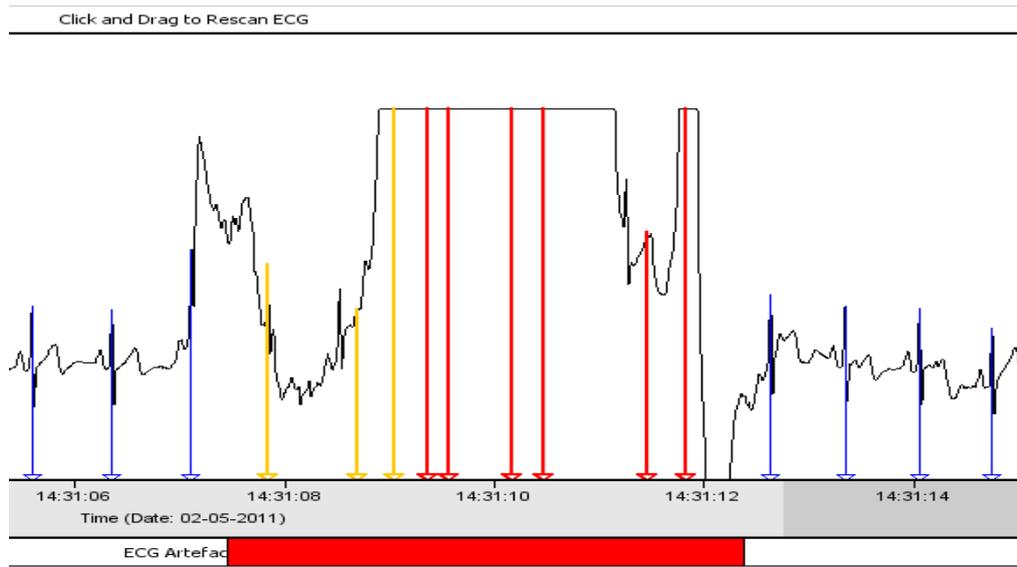
Step 2: R-peak detection

The *Detect R-Peaks* tab will assist you to create an artefact free IBI signal as fast as possible by applying automated artefact and peak detection. The mandatory visual inspection and correction of the resulting IBI signal is made as easy as possible by multiple zoom levels and an automated suspicious beat detector. You can also zoom in or out by using the scroll wheel of the mouse on the part of the data you want to see in more detail. By using the mouse wheel a shorter (scroll forward) or a longer (scroll backward) period of the recorded data can be shown.

The default settings of the R-peak detector work well on most of the ECG recordings but varies with signal quality. Automated artefact labeling reliably detects clipping and signal loss, but detection of noisy ECG is not perfect. Manual deletion of bad ECG signal parts may be additionally needed.

In the main window with the ECG signal the detected R-peaks are marked by vertical lines, mostly blue. A blue line means that the beat was considered to be correct according to the automatic beat detector. Potential mistakes in automated beat detection are termed 'suspicious beats' and are flagged by a red or yellow color. You can easily browse through all suspicious beats from most to least suspicious by pressing *Dot/right pointer* keyboard key for next, and *comma/left pointer* keyboard key for previous suspicious beat. Or you can simply use the menu buttons at the top of the window. When you are at a suspicious beat you can either delete a beat by right clicking on it, e.g. when a beat was placed in an obvious wrong location in between beats. Or you can add a beat by left clicking on the correct location of the R-peak, e.g. when a beat was completely missed. Just remember, suspicious is not always guilty.

To mark an unscorable / bad part of the ECG as an artefact, click with the left mouse button in the artefact bar and drag it from left to right until it covers the entire period that is to be marked as an artefact. All beats in the artefact period will be deleted from further analyses.



Assignment 2: Correct misplaced or missed beats and mark missed artefact periods.

Step 3: Segmenting / labeling the data

Here you divide the continuous data into logical periods for further analysis. We call this labeling. The aim of labeling is to get an average value for each available parameter, such as HR, PEP, and RSA, for each experimental condition or ambulatory activity.

Click on the *Label Data* tab. In the upper window we see the raw IBI time series and/or the smoothed (and hence lightly time lagged) heart rate signal. Because the heart rate signal is dependent on the quality of the IBI time series, make sure the R-peaks have been detected correctly in the *Detect R-Peaks* tab. In the lower window we see the ‘motility’ signal. This signal is based on the Y axis of the tri-axial accelerometer. The clock time and IBI time series are presented in the two bottom windows. Before we can actually begin labeling the data, we need to load the blueprint for all possible labels first. This blueprint lists all the experimental conditions or ambulatory activities. This is done in the label configuration file (.cfg file).

Assignment 3: load the label configuration file.

Go to the main menu – *Actions - Edit Label Configuration - Import Label Configuration From File*. Now select the *Morning Recording Rene.cfg* file and click *open*.

Assignment 4: label the activities from the activity diary

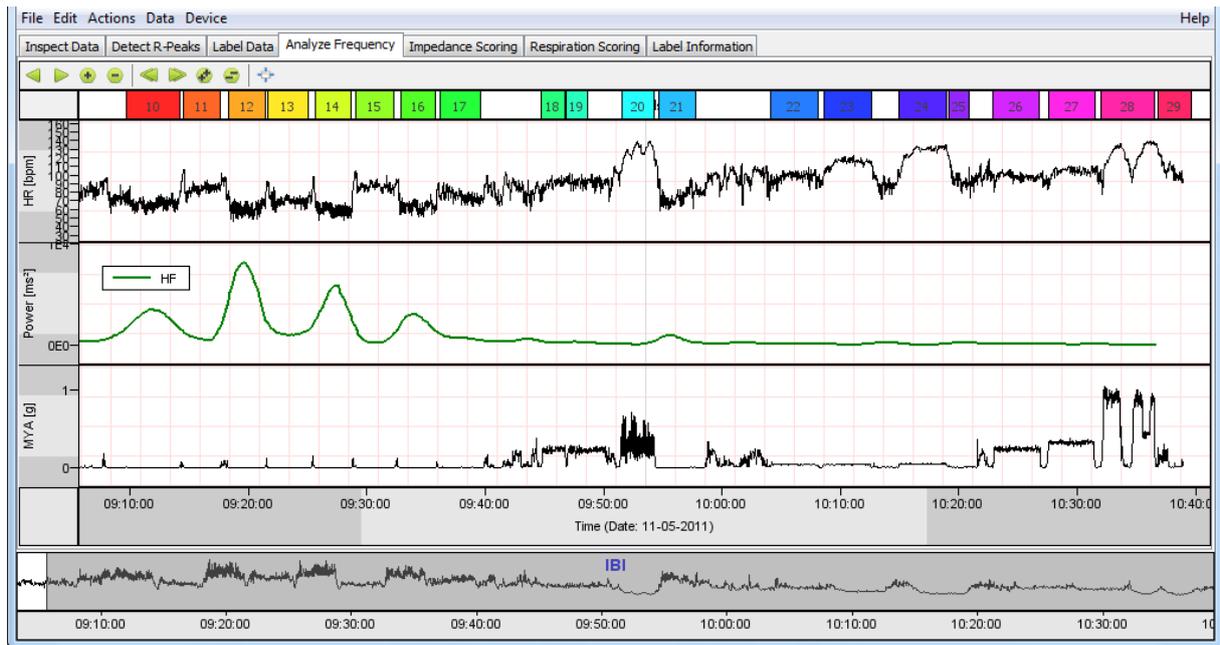
The label configuration is now loaded and now we can start the actual labeling of our data. Use the notes containing the start and stop times of each activity to draw labels in the top bar where it says “Click and Drag to add Labels”. When hovering the mouse on the bar you will see an indicator of the time window appear. This will help you place the starting point of your label accurately. Click with the left mouse button in the top bar at your starting point and drag the mouse to the right until the desired end time of the label is reached.

**A dashed vertical line will also appear on both ends of the label extending into the motility signal. This may help you remove the transition periods with movement between two experimental conditions from the final labeled data.*

***During the recording I pressed the event button of the VU-AMS device to mark the beginning and ending of each condition. These markers are displayed as lines here, and help us to more easily detect the start and stop times of each experimental condition. By clicking with right mouse button on the label bar between the two lines, we get the option to place a label between the lines automatically.*

Step 4: Spectral power calculations.

This is the most passive tab in DAMS. Spectral powers are calculated for every label with a label length above 4 minutes and there is no interaction possible. Let's move on!



Step 5: Impedance scoring.

The impedance cardiogram (ICG) is the first derivative of the change in thorax impedance using time as the basis (dZ/dt). This characteristic ICG waveform derives from the change in thorax impedance caused by left ventricular ejection of blood into the descending aorta during the systolic phase of the cardiac cycle. To improve signal quality, the ICG waveform is often obtained by ensemble averaging over beats within in a fixed time period, time locked to the R-wave peak. The typical period for ensemble averaging is one minute. In DAMS we deviate from this practice and instead compute a Large Scale Ensemble Average across the entire label (*see Riese et al., 2003 for the rationale*).

The most important variables extracted from the ICG is the preejection period (PEP). The PEP is an index of contractility which is only influenced by sympathetic but not parasympathetic activity in humans, which makes PEP the measure of choice to monitor changes in cardiac sympathetic activity non-invasively. The PEP is defined as the interval from the onset of left ventricular depolarization, reflected by the Q-wave onset in the ECG, to the opening of the aortic valve (B-point in the ICG).

Assignment 5: Score the ECG and ICG landmarks.

We will score all of these complexes together one by one to get familiar with moving around the impedance tab. The landmarks we will focus on are:

Q-wave onset in the ECG (the start of the electromechanical heart cycle).

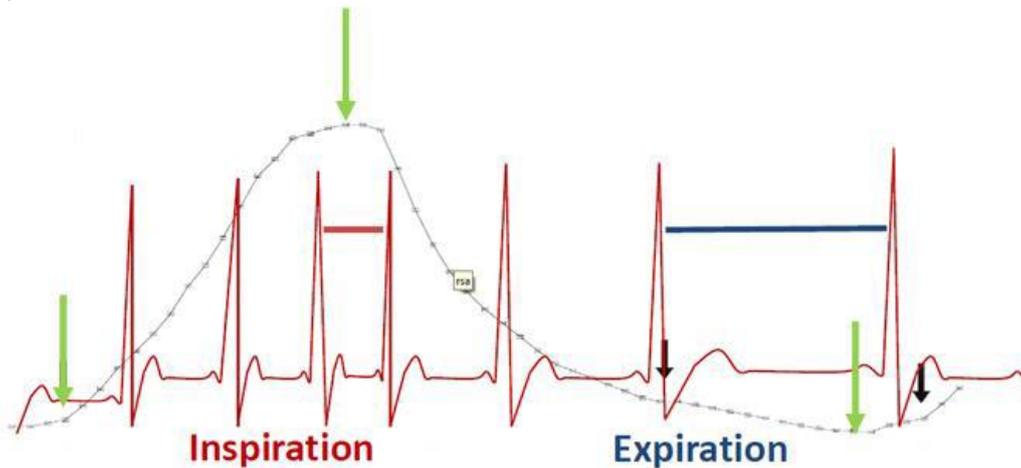
B-point or upstroke (the opening of the aortic valve)

dZ/dt min or C-point (max aortic diameter / velocity of aortic blood flow)

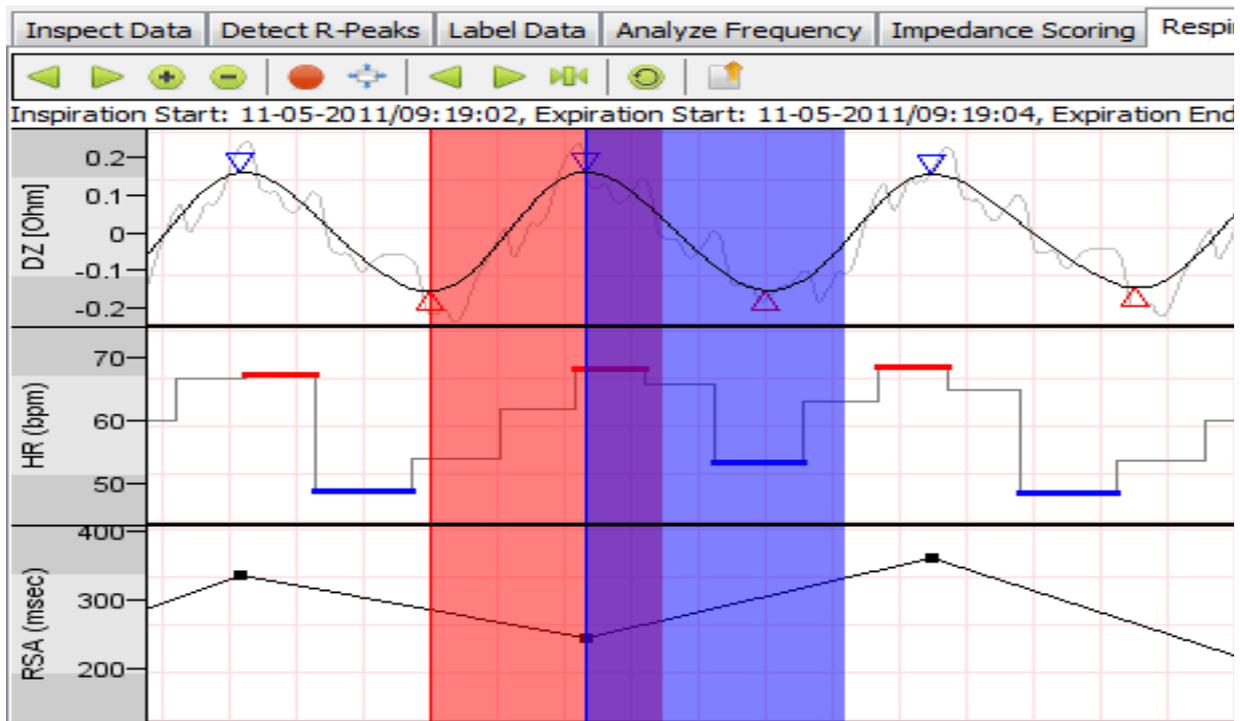
X-point or incisura (closing of the aortic valves = end of LVET)

Step 6: Visual inspection of the automated RSA detection.

Respiratory Sinus Arrhythmia (RSA) scoring by the DAMS program is based on the peak-valley method (Grossman, van Beek, & Wientjes, 1990 ; de Geus et al., 1995) that uses the IBI time series extracted from the ECG together with the respiration signal obtained from filtered (0.1 – 0.4 Hz) dZ signal to obtain heart period variability that is associated with respiration. This heart period variability is referred to as RSA.



The DAMS program contains an automatic scoring algorithm for detecting the beginning and end of inspiratory and expiratory phases in each respiratory cycle. For each respiratory cycle the total cycle time between begin of inspiration and end of expiration is extrapolated to a per-minute respiration rate (RR). In addition, RSA is computed per respiratory cycle from two IBIs: The shortest IBI during an interval starting at the begin of inspiration and ending 1000 msec (default) after the end of inspiration and the longest IBI during an interval starting at the beginning of expiration and ending 1000 msec (default) delay after the end of expiration. RSA is calculated by the subtraction of the shortest IBI from the longest IBI, provided that the shortest IBI (highest HR) is part of an accelerating series within the inspiratory interval and the longest IBI (lowest HR) of a decelerating series within the expiratory interval. This is illustrated in the figure below.



If either the decelerating longest or accelerating shortest IBI is missing for a breath cycle, or a negative RSA value is obtained on subtraction, we set RSA in these breaths as missing.

Assignment 6: Visually inspect the RSA scoring

Automated scoring of the respiration signal works quite well in most subjects. Mostly, it will suffice to just load the .amsdata file into DAMS and browse through the signal after having set the time axis at a low temporal resolution (e.g. ten minutes per screen). While browsing through the respiration signal from the beginning to the end of the file check the following:

Clipping dZ : Purpose: The purpose of this option is to automatically reject clipping (i.e. where the raw respiration signal turns into a flat line at $dZ = 1$ ohm or $dZ = -1$ ohm). The default values usually suffice and detected true clipping very well. Just check if it is true clipping when more than 10% of the recording is marked as a clipping artefact.

Irregular respiration : Do all inspirations and expirations appear to be appropriately scored in the upper respiration signal (indicated by blue and red triangles)? --> If erroneous breaths are scored, did the program mark them as artefacts in the bar at the bottom of the screen labeled "Irregular respiration"? --> Add or remove irregular respiration artefacts where needed.

**Did the program mark more than 10% of the recording as an "Irregular respiration" artefact If so, the parameters of the scoring algorithm may need to be changed under settings in the menu bar.*

Irregular IBI: The purpose of this option is to automatically reject spikes in the IBI time series, that represent extra-systolic beats or very prolonged beats. --> Check whether the program has rejected all deviant IBIs (spikes) without removing IBIs that reflect large but true heart rate variability. Hence make sure all spikes are marked as artefacts in the bar at the bottom of the screen labeled "Irregular IBI".

**The difference between a spike (e.g. caused by premature ventricular contraction (PVC) and a truly high heart rate variability is rapidly gleaned from the shape of the tachocardiogram. If there is a staircase pattern rather than a sudden single-beat change, the subject may have a generally high*

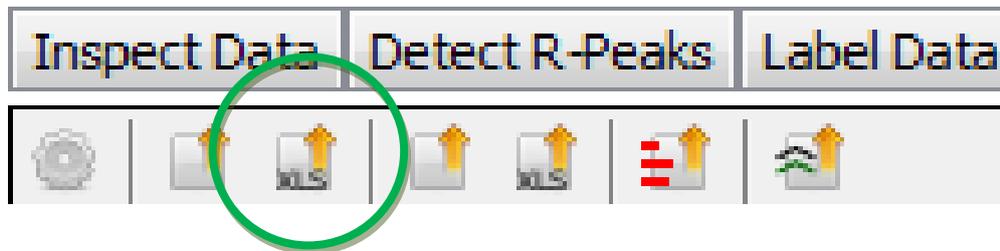
heart rate variability which can be verified in the RSA window (e.g. RSA > 200 msec). If there is a sudden single-beat drop or jump then there is a spike. Spikes often represent extra-systolic beats or very delayed beats (which often occur jointly).

Step 7: Exporting the results

After clicking on the *Label Information* tab wait until all cells are finished calculating. The first column in the *Label Information* tab is the subject name, which is the ID that we gave when programming the device for recording. Each row represents a single labeled experimental/ambulatory condition and each consecutive labeled condition is rank-ordered by the *Label ID* field. The rest of the columns contain values for a large number of physiological variables. ICG-based variables only have a value when the scoring under the *Impedance Scoring* tab has been done (e.g. PEP) and the LF and HF power values from spectral analyses on the IBI time series are only present for labels with a minimum length of 4 minutes.

Assignment 7: Export the results to ASCII

Click on the export button in the menu and you will be prompted to enter the location to the output file.



More practice: PEP and RSA scoring

You will find two folders on the hard drive containing the following:

`G:\SAA workshop 2013\EXPERIMENTAL AMS DATA\Yourname\`

Every folder contains several AMS files and an SPSS script that we will later use for initial read-in and merging of your data.

Assignment 8: Inspect the impedance scoring of each recording

Inspect the impedance scoring of each recording and export the output to a single ASCII file in your directory. Here is a summary of the PEP scoring rules that help you go through the complexes:

Morphology

The B-point or upstroke should be at a first or second order zero-crossing in the dZ/dt signal. It should be close to the $dZ/dt=0$ line, and be the starting point of the longest uphill slope before the dZ/dt_{min} point. However, rather than appearing as a clear incisura, the B-point may sometimes take the form of a subtle inflexion and may vary considerably from beat to beat. It is therefore very important to inspect the dZ/dt signal closely in order to identify it.

The dZ/dt_{min} is normally visible as a clear peak in the window between the B- and the X-point. In some cases the dZ/dt signal shows a double peak, a bit like rabbit ears. If one of the peaks is clearly (40%) higher than this peak is chosen. If the peaks are of comparable magnitude, choose the first peak.

The X-point or incisura is always a local minimum after the dZ/dt_{min} . Often it is the lowest point in the entire signal, but not necessarily. In the ideal situation it can be seen as a sharp trough in the ICG signal. This is the most clearly identifiable choice for the X-point. It may be that two or more troughs lie in close proximity without one being clearly the lowest point in all complexes. The latter part of the ICG waveform then looks like a "W". In this case choose the second trough (mostly, this trough is usually followed by the longest uphill slope after the dZ/dt_{min} point).

Consistency

Whatever point you choose, choose that point consistently. If a "less-than-ideal" upstroke is present in all complexes, but an "ideal" upstroke is present in some, choose the less-than-ideal one in all complexes, even those featuring a more "ideal" upstroke. Before starting to score the ICG, try browsing through the entire ICG signal first. You can then decide which points can be most consistently identified, and this holds for both for the B-point and the X-point.

Physiological plausibility

We can use the ECG signal to make decisions a bit easier. e.g. the B point will probably not take place around the Q-onset or T-top. Below are some plausible values for each interval based on recordings in healthy adults.

HR: 40-60 → PEP: 100-140 → LVET 300-450

HR: 60-80 → PEP: 90-130 → LVET 250-400

HR: 80-100 → PEP: 80-120 → LVET 200-350

HR: 100-120 → PEP: 70-100 → LVET 200-300

HR: 120+ → PEP: < 80 → LVET 150-250

Again, if your signal shows B- and X-points outside of these ranges, this does not at all mean that your dZ/dt signals should be discarded. The above table is just a general rule of thumb.

** As you will notice some recordings need quite a lot of zooming to see the Q-onset or have no Q-onset no detectable point. We set the Q-onset to missing across all complexes in these latter cases. The PEP will then be calculated from R-B interval + a fixed interval based on the average of a large population of normatives.*

Multiple Rater comparison

Reliability increases if two (or more) raters score the same data set independently. After inter-rater reliability is established, the various raters should ideally compare their deviant scoring to converge on a single solution, in view of the consistency principle. Mostly one will have picked a different B-point then the other(s). Averaging the B-point location is meaningless. Consensus has to be reached on the correct B-point location to satisfy the consistency criteria.

Assignment 9: merge the output ASCII files in SPSS

Run the script with your name in the title to merge all output files.

Assignment 10: inter rater reliability

Appendix III

Run the script mentioned "merging_" in pairs (pre-defined buy your coupled names) and compare the within subject correlations. Where did you differ in choice and is there a consensus to be reached?