A SYSTEM FOR CONTROL AND EVALUATION OF ACOUSTIC STARTLE RESPONSES OF ANIMALS

Z. Bureš

College of Polytechnics, Jihlava
Institute of Experimental Medicine, Academy of Sciences of the Czech Republic, Prague

Abstract

Psychophysical experiments on animals often use behavioral tests to assess various aspects of the animal's perception. One of the behavioral methods employs the startle response, which represents an unconditioned response to an audible, tactile or other sensory stimulus. The acoustic startle response (ASR) is a reflex reaction elicited by loud auditory stimuli. The ASR amplitude can be altered when the startling stimulus is immediately preceded by another stimulus (prepulse). A MATLAB tool is presented that serves as an environment for controlling the experimental procedure, generating signals, and for basic evaluation of the ASR.

1   Psychoacoustic measurements in animals

Psychophysical experiments on animals are often conducted to determine the animal's subjective perception properties. One of the possible methods of such experiments is animal training that develops a conditioned response. A classical scenario, for example, is the experimental procedure to assess the animal's hearing threshold. In this case, the animal is trained to stop drinking whenever it perceives a sound, thus the perception of the stimulus may be derived based on the conditioned behavior. However, animal training is a demanding and time consuming task; for this reason, other methods were sought that would avoid the training procedure. One of these methods is based on startle reaction – a transient motor response to an intense unexpected sensory stimulus. Startling is a primitive unconditioned behavior observed in all mammals. In spite of its relative simplicity, the startle reflex shows several forms of behavioral plasticity including habituation, sensitization, and prepulse inhibition [1]. In the case of auditory psychophysics, the startling stimulus is a loud noise or tonal stimulus, the reaction is then called the acoustic startle reflex (ASR). The ASR may be used as an indicator of the behavioral responsiveness to suprathreshold sound stimuli. An example of a dependence of startle amplitude on sound intensity is shown in Fig. 1.

From its very principle, the startle reaction can not be used directly for measurement of, e.g., hearing thresholds – the ASR is elicited only by intense stimuli. However, the ASR amplitude can be altered when the startling stimulus is immediately preceded by another stimulus (prepulse). In this case, the startle reaction can be either enhanced (prepulse facilitation) or suppressed (prepulse inhibition, PPI), compared to the case in which the startling stimulus is presented alone. Using this experimental procedure, audibility of a prepulse sound may be assessed on the basis of alteration (inhibition) of the startle reaction.

The basic experimental procedure for the ASR measurement is as follows: startling sounds of varying intensity are generated and presented to the animal in random order and with random intertrial intervals. The animal is placed in a small chamber on a sensitive platform that detects the animal's movements. The amplitude of the movements is subsequently evaluated. The procedure for the measurement of the prepulse inhibition (PPI) of ASR is analogous, the difference is that the startling stimulus remains constant during all trials, while the prepulse sound varies. The interval between the prepulse and the startle stimulus is fixed across all trials. A system for control of the experimental procedure must provide the above mentioned functioning and be capable of a flexible experiment design. As the software system supplied with the behavioral measurement apparatus (Coulbourn Habitest, model E10-21 [2]) was found unusable due to numerous severe limitations, a custom system was implemented in MATLAB environment to fulfill all the requirements. The tool is equipped with a graphical user interface.
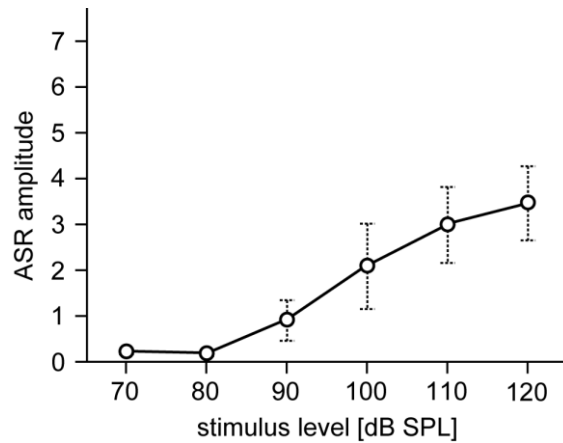
Figure 1: Example of an averaged ASR response of a rat in dependence of the startle stimulus level. Mean values ± SEM are shown.

2   Experimental setup

The entire system for measurement of acoustic startle responses comprises several components (see Fig. 2). A computer with the presented MATLAB tool is used for control of the experiment, it provides generation of the stimuli, recording of the responses, as well as basic statistic evaluation. From the PC, the stimulus waveforms sampled at 50 or 100 kHz are fed into a TDT system III with Real-Time Processor RP 2 (Tucker Davis Technologies [3]) equipped with analogue output. The analogue waveforms are lead into a custom made amplifier and a speaker placed inside a startle response chamber (Coulbourn Habitest, model E10-21). The animal's movements are detected by a sensitive platform (Coulbourn Habitest, model E45-11) and converted to differential voltage which is preamplified and processed by a controller (Coulbourn Habitest, model A10-21). The resulting voltage is sampled using the TDT system and sent to the PC in a digitized form.
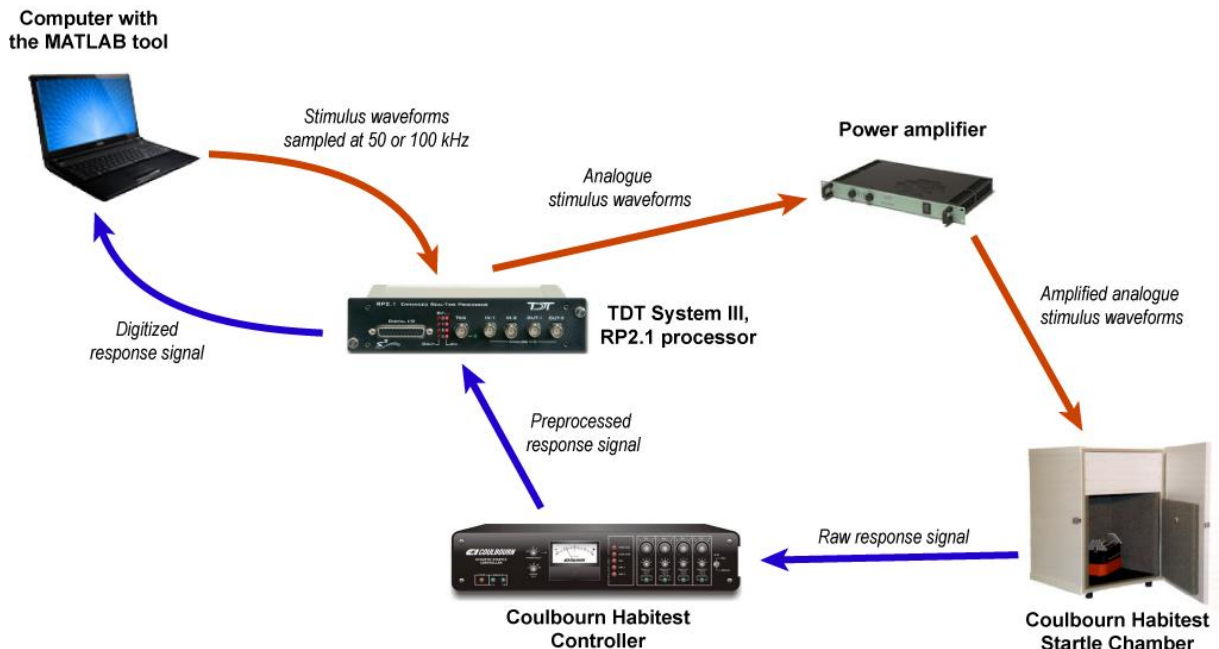

Figure 2: Experimental setup used for the ASR measurement.

3 Graphical user interface and basic fuctions

At the beginning of the experiment, several global parameters must be set, such as the sampling frequency, length of the stimulus ramp, length of the recorded responses, or background noise. These parameters are grouped at the top of the graphical user interface (GUI). The main window of the GUI is shown in Fig. 3.

Each experiment consists of many trials with different stimulus parameters. Each trial can be individually designed in upper region of the GUI. Parameters of the prepulse (denoted as SIGNAL in the GUI) and of the startle stimulus may be set individually, it is possible to choose between a tone with a given frequency, white noise, and an arbitrary waveform loaded from a WAV file. For each sound, its intensity and duration may be set. Additionally, the delay between the prepulse and startle stimuli may be set. A prepared trial is subsequently inserted into the experimental scenario (denoted as Script in the GUI), either at cursor position (INSERT button) or at the end of the script (APPEND button). It is also possible to load the parameters of a selected trial from the script back to the input fields and to modify and update the trial parameters (UPDATE button).

Any psychophysical experiment requires repeated presentation of the same trial to enable statistical evaluation of the probability of correct answer. To support this requirement, a selected trial from the script may be easily duplicated for a given number of times (DUPLICATE button).

When all trials are prepared and inserted into the scenario, the order of trials may be randomized and interleaved with random inter-trial intervals (denoted as ITI in the GUI). Trials can also be moved up and down in the scenario, or deleted. Scripts may be saved to a file for future repeated experiments.

The experiment is started off with the RUN button. It is possible to stop the experiment at any time. The responses to each trial are recorded and showed immediately in the response graphs. Individual response waveform (upper graph) as well as extracted response magnitudes (lower graph) are shown. The results of an experiment may be saved to a file to allow subsequent processing.
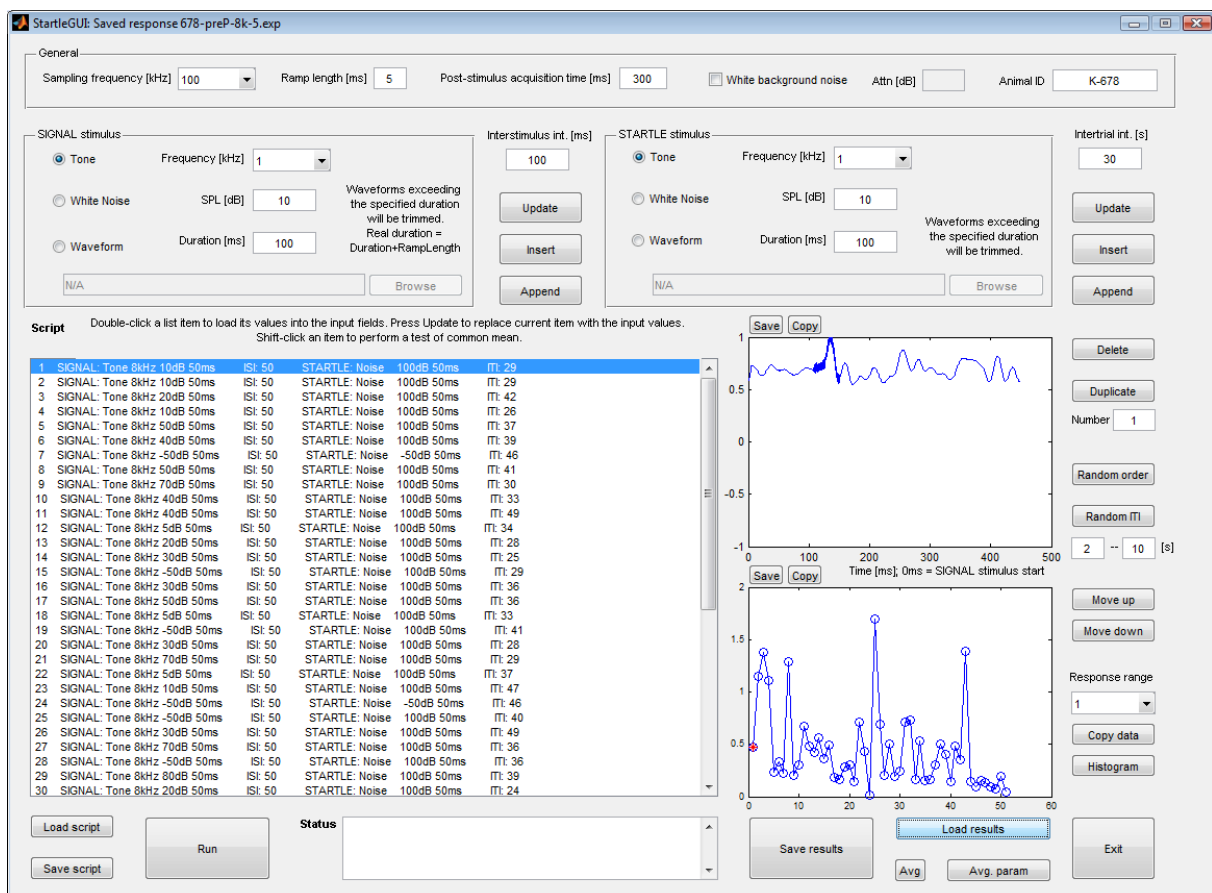

Figure 3: Graphical user interface, individual trials. The entire experimental scenario is shown along with the response waveform of the trial under the cursor (top-right graph) and extracted response amplitudes of all trials (bottom-right graph).

Basic offline evaluation of the experimental results is available. The results may be loaded and analyzed by individual trials (LOAD RESULTS button), in this case, unwanted trials may be deleted and the results may be saved under a diferent filename. Another possibility is to group the identical trials together and compute mean response amplitudes for each trial type (AVG. PARAM button). In this case, the individual response waveforms are not shown; instead, an averaged amplitude-trial function is shown in the lower graph in the form of mean and variance of amplitude for each trial type (see Fig. 4). To order the trials according to a chosen parameter (e.g., prepulse level), it is possible to move the trial types up and down in the scenario; the graph is updated accordingly. Basic statistic tests for testing the mean difference between two trial types is also available. A histogram of response amplitudes can be shown for any trial type.

Both result graphs may be copied to clipboard to enable easy presentation of the data. Furthermore, all kinds of results may be saved to a file in textual form to enable subsequent processing.
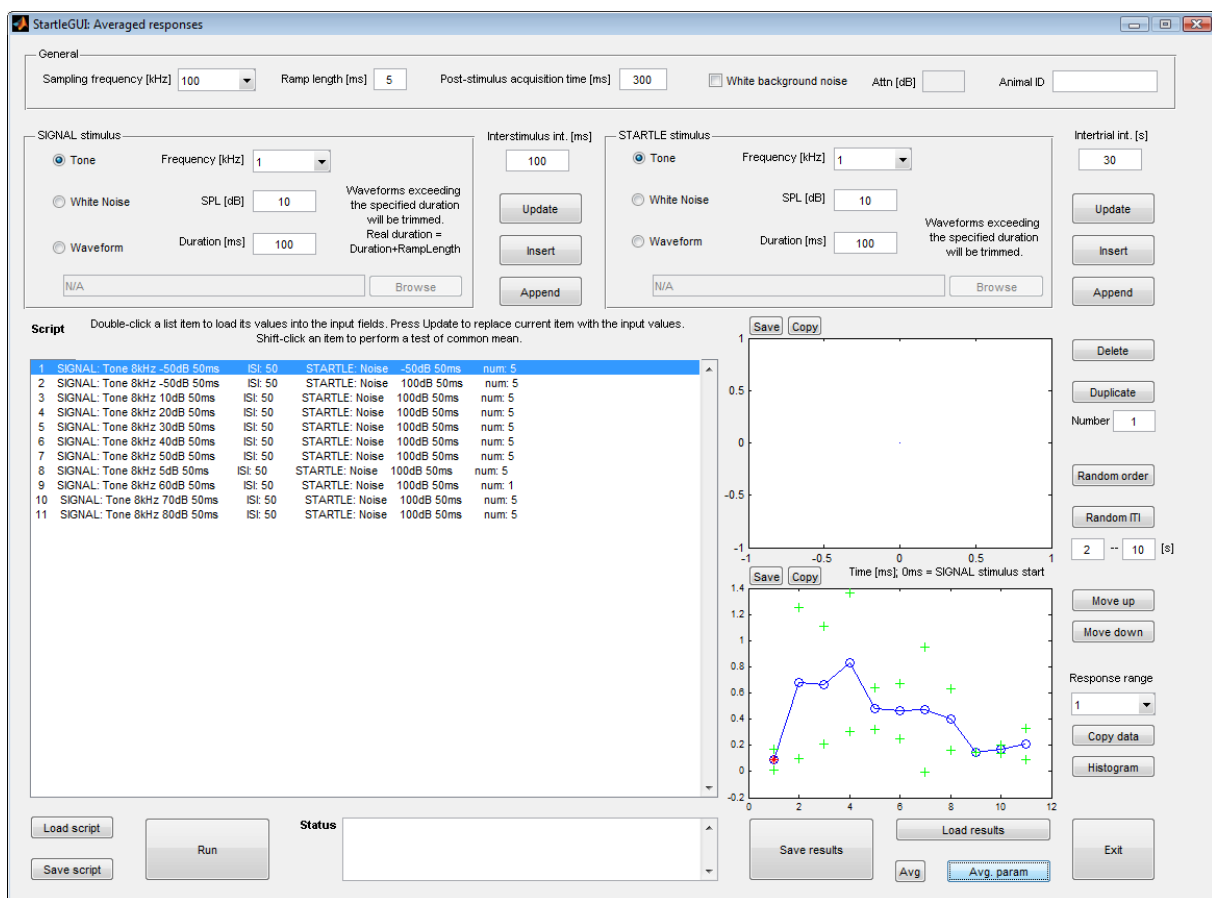

Figure 4: Graphical user interface, averaged evaluation of results. Identical trials are grouped together, the botton graphs shows mean and variance of response amplitude for each trial type.

4   Conclusions

The presented MATLAB tool represents a custom software solution that fulfills all the requirements on behavioral experiments employing startle responses given by routine practice at the Department of Auditory Neuroscience at the Institute of Experimental Medicine of AS CR. The system replaces the unusable software system supplied with the experimental apparatus (Coulbourn Habitest). Thanks to several ways of data visualization, the experimenter is able to evaluate the preliminary results immediately. Export of both textual and graphic data enables further processing and easy presentation of the results.

References

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Ing. Zbyněk Bureš, Ph.D. [1,2]
[1] Dept. of Electrotechnics and Informatics, College Of Polytechnics Jihlava
Tolstého 16, 58601 Jihlava
buresz@vspj.cz
[2] Institute of Experimental Medicine, AS CR
Vídeňská 1083, 14220 Praha 4
buresz@biomed.cas.cz