

Video Article

Psychophysically-anchored, Robust Thresholding in Studying Pain-related Lateralization of Oscillatory Prestimulus Activity

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Abstract

In perceptual studies, it is often important to objectively assess the equality of delivered stimulation across participants or to quantify the intra-individual sensation magnitude that is evoked by stimulation over multiple trials. This requires a robust mapping of stimulus magnitude to perceived intensity and is commonly achieved by psychophysical estimation methods such as the staircase procedure. Newer, more efficient procedures like the QUEST algorithm fit a psychophysical function to the data in real time while at the same time maximizing the efficiency of data collection. A robust estimate of the threshold intensity between painful and nonpainful perceptions can then be used to reduce the influence of variations in sensory input in subsequent analyses of oscillatory brain activity. By stimulating at a constant threshold intensity determined by an adaptive estimation procedure, the variance in the ratings can be directly attributed to perceptual processes. Oscillatory activity can then be contrasted between "pain" and "no-pain" trials directly, yielding activity that closely relates to perceptual classification processes in nociception.

Video Link

The video component of this article can be found at <https://www.jove.com/video/55228/>

Introduction

When conducting behavioral experiments involving human participants, it is important to be able to closely control the intensities of presented stimuli. Using stimuli of equal intensity for all participants, however, will in some settings introduce the bias of subjective perception. For some perceptual qualities such as pain, there are high inter- and intra-individual variations in perceived intensity at a constant stimulus level^{1,2}. For experiments that assume equal subjective percepts, it is thus a necessity to match the subjectively perceived intensity across participants. This is also important when examining perception at threshold level, e.g., between painful and nonpainful stimulation. Psychophysics research has addressed these kind of problems for decades, and today there are sophisticated but easy-to-use methods available to achieve robust psychophysical anchoring.

A simple, classical method of mapping the intensity of a stimulus to an individual sensation magnitude is the staircase method³. Hereby, the intensity of successive stimuli is increased or decreased, until there is a change in the participant's response relating to the desired threshold or position on the subjective sensation scale. Repeating this process number of times, yields a plausible estimate of the reversal point. Classical methods, however, fail to make use of all the information contained in each rating trial. This leads to an unnecessarily high number of trials required to reach convergence. Methods such as (linear) regression or function fitting might fail, if the assumptions for the relationship between stimulus intensity and sensation magnitude are wrong or do not hold for the tested stimulus range. The adaptive procedures not only yield a robust point estimate for a certain subjective intensity, but do so more efficiently. Especially for longer experiments, which heavily rely on accurate estimation of a threshold or sensation magnitude, it is necessary for the psychophysical method to be both robust and at the same time efficient with respect to the number of required trials. This is especially important in fields such as pain research, where the total exposure to painful stimulation should be kept as low as possible for the participants' benefit.

Although the classical staircase methods are still widely used, e.g. in quantitative sensory testing, the use of more advanced estimation methods that make better use of the acquired information across trials is steadily increasing. In the case of the maximum likelihood estimation method QUEST^{4,5} used here, this is probably due to the readily available implementation in the popular Matlab PsychToolbox⁶ suite. The modern, revised version of this procedure is superior to classical estimation methods both in robustness and the low number of trials required to arrive at a sufficient estimate, if used with the right settings⁷.

The rationale behind the QUEST procedure is to fit a Weibull function to the incoming data to model the psychophysical transformation between stimulus intensity and sensation magnitude. The parameters for the psychophysical Weibull function are in part given by the experimenter, e.g. the steepness of the function or the offset due to the false positive rate and responder inconsistency. The positioning of the parameter of interest along the intensity dimension is approximated by the procedure using Bayesian maximum likelihood estimation. Hereby, a probability distribution is assumed over the location of the target parameter, i.e. the threshold intensity. Given a sensible prior assumption for such a distribution, the

algorithm will determine the most informative intensity that the participant should respond to. For the current implementation of the procedure, this is the mean of the prior probability distribution⁸. For each successive trial, the prior probability distribution is in essence multiplied with the likelihood of the participant's given response at the tested stimulation level, as characterized by the Weibull function. Every response will be used to continuously update the probability distribution estimate for the threshold parameter. This procedure is repeated until a satisfying estimate is produced. The procedure is more efficient than a simple regression because it makes immediate use of the collected responses to determine which stimulation intensity to test next. Also, the procedure will specifically probe around the point of interest, e.g. a threshold or certain sensation intensity. Using only testing data from such a limited range in regression would lead to an unstable estimate, making adaptive procedures more robust in settings where only relatively low numbers of trials are feasible.

Such robust psychophysical anchoring can be used to measure changes in pain sensitivity over time, modulatory effects in hyperalgesia/allodynia research or analgesic effects in pharmacological interventions, amongst other settings. Another interesting prospect of being able to anchor stimuli to the intensity just at the threshold between two sensory continua is to examine subjective perception across the transition from non-painful to painful sensation^{9,10,11}. This scenario is very interesting because if the pain threshold has been robustly estimated, pain and no-pain conditions can be contrasted in electroencephalographic (EEG) activity, for example, without changing the physical stimulus intensity¹². This allows for the observation of pain-specific perceptual processes under constant stimulus conditions by examining the difference in brain activity between trials rated as painful and non-painful.

We will demonstrate how to use the readily available implementation of adaptive estimation in PsychToolbox to robustly determine the individual pain threshold in an EEG experiment where the contrast between pain and no-pain activity is examined for lateralization effects, depending on the stimulation site. Since the stimulation intensity can be kept constant after the thresholding procedure, it is not necessary to account for EEG activity co-varying with stimulus intensity in the subsequent analysis.

Protocol

The experiment has been approved by the ethics commission of the Hamburg medical association (PV4509).

1. Participant Selection

1. Beyond standard selection criteria, such as fitness for pain stimulation, head implants or pre-existing neurological conditions, make sure the participants are not suffering from acute or chronic pain, are not taking any pain medication, and have no known history of substance abuse. Participants should also not have taken part in any pharmacological studies during the 4 weeks prior to the experiment.
2. Include participants of any gender, yet take care to only include female participants that are using hormonal contraceptives^{13,14} to minimize the effect of cyclic changes in pain perception.
3. Before administering any kind of stimulation, make sure participants have given informed consent in writing.

2. EEG Setup

1. Select an appropriate cap size and prepare the EEG electrode setup as per the system's instruction manual.
2. Set the sample rate and high/low cutoff as well as the impedance limits of the recording equipment (recommended: 500 Hz, 0.5 Hz high-pass filter, impedances <20 kΩ).
3. Make sure the stimulation device and the EEG device are not electrically coupled by running the EEG system on battery.
4. Ensure that any link between the EEG system and the computer controlling the electrical stimulation device is potential-free.

3. Electrical Stimulation Setup

1. To best make use of the time resolution of the EEG recording, keep the electrical stimulation as short as possible. Set the stimulator to a single, monophasic stimulation pulse with 1 ms duration and 400 V maximum voltage. If a more intense pain level is needed, or the exact timing of the post-stimulus EEG recording does not take precedence, other stimulation protocols can be used.
2. Make sure the electrical stimulator is switched on but the output to the electrode is switched off. For the DS7A stimulator the switch labeled 'OUTPUT' to the right of the device should be in the down position.
3. Locate the landmark(s) that identify the chosen stimulation site. For a stimulation at the hand, use the muscle between thumb and index finger (abductor/flexor pollicis brevis). Ask the participant to lay their hand on a flat surface with all digits stretched out and apposed. Identify the stimulation site by bisecting the distance between the first knuckles of the thumb and index finger.
4. Clean the skin by applying electrode preparation gel. Make sure not to use alcohol or disinfectant, which might leave residue on the skin that can lead to irritation or unreliable stimulation.
5. Attach the stimulation electrode and fasten it in place with textile tape.
6. Ask the participant to find a comfortable position for the hand and to try not to move the hand during the experiment, if possible. For the participant's convenience, place a soft tissue under the hand to absorb any humidity, depending on the surface permeability.
7. Enable the stimulator's output by switching the 'OUTPUT' switch to the upward position.

4. Determine Starting Points

1. Instruct the participant on how to operate the rating scale on the screen using the mouse. The left half represents non-painful sensations; the right half corresponds to a standard pain VAS scale, providing a visual equivalent for a range of continuous sensation intensities in the form of a horizontal line. Point out to the participant that the absolute center point of the scale cannot be selected. Provide the participant with the standardized instructions about the anchor points¹⁵ (Table 1).

2. Give the participant the opportunity to get comfortable with the rating process by applying stimuli of varying intensity and recording the responses. Use the information gathered during this phase to get an estimate for two intensities that consistently evoke strong but nonpainful sensations (low-point) and moderately painful sensations (high-point), respectively. Continue the stimulation for about 25 - 30 trials or until satisfied with having reached good estimates. During this time, it can be beneficial to query the participant for verbal feedback on the intensities and the subjective similarity of repeatedly presented stimulus intensities.
3. Try to pick the intensities randomly to evoke responses around the scale center. For best results, do not simply increase or decrease the intensities linearly, and also explore the more extreme ends of the painful side. This phase should also give the participant the opportunity to get accustomed to the potentially unfamiliar stimulation and establish some reference for a consistent rating range. Because of this, it is advisable to apply intensities from the whole range of possible stimulus intensities, while also repeating some intensities.
4. Once satisfied with having obtained estimates for both a high-point and low-point starting intensity, inform the participant that the first part of the experiment is about to start and (s)he should keep on rating as practiced while random stimulus intensities are presented.

5. Determine Threshold

NOTE: The QUEST algorithm requires some parameters to be specified before starting estimation. Those parameters include the steepness of the psychophysical function (beta, typically 3.5), the fraction of trials where a random answer is expected (delta, typically 0.01), and the fraction of trials where a positive response is expected even though no stimulation is given (gamma, no recommendation). For Bayesian estimation, the range (SD) of the expected ratings and the spacing of possible responses (grain) must be specified. For a VAS, grain should be set to the resolution of the scale (typically 1), and the SD should be set large enough to include both the scale zero point and the maximum possible intensity plus some safety margin. The recommendations and "typical" values given here are explained in detail in the QuestCreate source code included with PsychToolbox^{6,16}. For pain at the threshold, a gamma value of at most 0.01 should be plausible. The estimation method is relatively robust in terms of misspecification of the parameters, however for settings with only few trials, failure to specify sensible parameters might increase the uncertainty of the final estimate. If the standard deviation is set too low, the procedure will have problems converging on estimates that lie outside the area spanned by the standard deviation around the prior guess for the parameter. Thus it is important to rather err on the side of a too large standard deviation.

1. Create two QUEST sessions with the parameters given above. Start one from the high-point intensity and one from the low-point. Information about the implementation logic of the estimation process can be found in the supplemental material (S1).
2. Randomly select a probe intensity of one of the two runs given by the respective QuestMean function.
3. Set the electrical stimulator to the probe intensity. If a different intensity than the one suggested by the algorithm needs to be applied or the suggested intensity is out of range, feed the presented intensity back into the QuestUpdate function in step 5.5.
4. Trigger the stimulus.
5. After the participant has rated the stimulus, run QuestUpdate for the respective estimation session and supply it with the actual stimulus intensity presented as well as the participant's rating.
6. Continue running rating trials until the estimates are stable or a predefined stopping criterion (>40 trials) has been reached.
7. Record the mean threshold estimate between both estimation runs, starting from the high and low starting point as given by QuestMean.
8. Allow the participant to take a break at this stage, if desired.

6. Stimulate at Threshold Level

Note: It is possible to adjust the rating and block count to your needs as long as it is tolerable to the participant.

1. Inform the participant that for the remaining part of the experiment, more blocks with random stimulation will be following and they should keep rating as they did before. If needed, refresh the instruction on the scale anchor points.
2. Start the EEG recording.
3. Set the electrical stimulator to the mean threshold estimate obtained in step 5.7 and keep the setting constant throughout the rest of the experiment.
4. Start a rating block (30 trials) and observe the data quality of the EEG recording. Depending on the EEG data quality, run 4 - 5 rating blocks and allow the participant to take short breaks in between the blocks.
NOTE: Try to keep social interaction with the participant to a minimum during these breaks or standardize the interaction as much as possible.
5. When finished, stop the EEG recording, switch the stimulator output to off, and remove the electrode. Debrief the participant after removing the EEG cap.

Representative Results

Using a rating scale split into one half for nonpainful and one half for painful sensations (**Figure 1a**), constant stimulation can be applied over many trials while still yielding ratings across the scale midpoint (**Figure 1b**). This way, changes in sensory input can be avoided, and the rating outcome can be directly related to intrinsic perceptual classification processes related to pain.

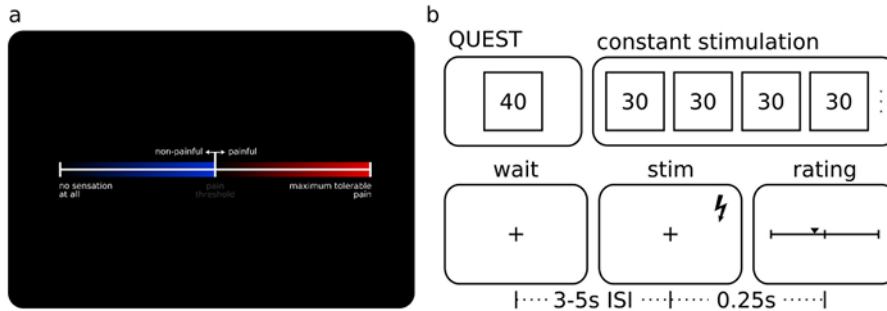


Figure 1: Experimental Description. (a) The rating scale with the left side spanning non-painful sensation and the right side spanning painful sensation. (b) Procedure used for data collection. 40 thresholding trials followed by 4 - 6 blocks of constant stimulation (30 trials each). The blocks had a jittered 3 - 5 s intertrial interval (ITI). The rating scale appeared 0.25 s after stimulation. [Please click here to view a larger version of this figure.](#)

The two estimation runs starting from the "nonpain" low-point and "pain" high-point converge on robust threshold estimates. Taking the mean of both estimates yields the final threshold estimate, while the bias induced by the starting intensity is reduced (**Figure 2a**). The subjective stimulation intensity evoked by repeated stimulation at the estimated threshold is stable across multiple blocks of 30 trials each within one experimental session (**Figure 2b**).

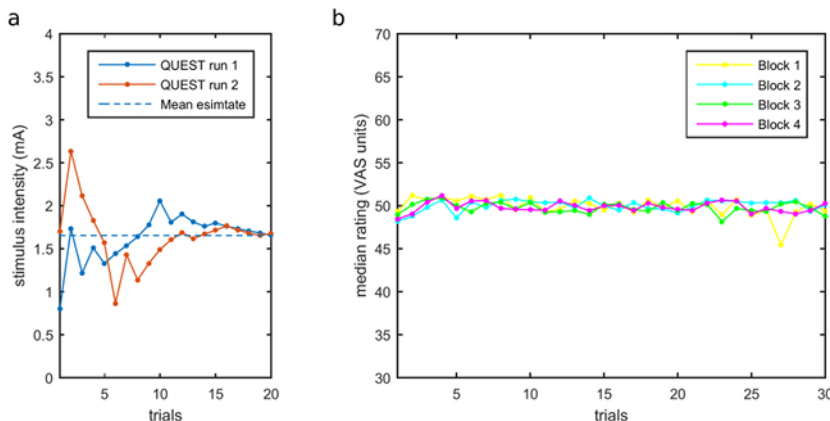


Figure 2: Stability of the Threshold Estimates. (a) Data for a single participant showing the algorithm converging on two estimates, one for a high intensity starting point, one for a low intensity starting point. To minimize the influence of the starting point, both threshold estimates were averaged (dashed line). (b) Stability of the rating medians over the course of the experiment under constant stimulation at the estimated threshold across all participants ($n=25$). [Please click here to view a larger version of this figure.](#)

By splitting the concurrently recorded EEG data into trials that were rated as "painful" and "nonpainful", respectively, the oscillatory activity can be contrasted post-hoc. This yields activity differences which coincide with perceptual decisions about the same stimulus being categorized as strong sensation or as pain. **Figures 3a** and **b** show these differences for a time-window before the painful stimulus is presented (-0.8 s to 0 s before stimulus onset) and the theta-band frequency range ($4 - 7$ Hz), which have previously been shown to be connected to subsequent perceptual classification in pain¹⁷. The thresholding paradigm enables the examination of such prestimulus differences in oscillatory activity linked to subsequent perceptual classification of pain, independent of stimulus magnitude.

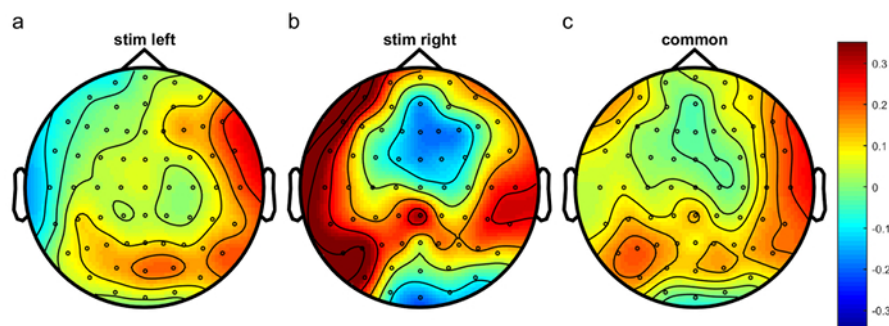


Figure 3: Power Differences between Nonpain and Pain. Data has been transformed to time-frequency domain using a multi-taper method. Depicted are Theta frequencies between 4 - 7 Hz and before stimulus onset (-0.8 s - 0 s). **(a)** Power difference specific to the subsequent classification of the stimulus to the left hand as painful. Data adopted from Taesler & Rose¹⁷ (n = 15). **(b)** Power specific to classification of a stimulus to the right hand as painful (n = 10). **(c)** Common Theta activity between (a) and (b), independent of the stimulated side (n = 25). The topo-plot shows the sum of the lateralized differences between painful and non-painful stimulation. For individual pain/no-pain topographies (S2) as well as a comparison to pre-existing post-stimulus data¹⁰ (S3) please refer to the supplemental materials. [Please click here to view a larger version of this figure.](#)

By changing the side of the stimulation between groups, these pre-stimulus effects can further be disentangled from any lateralization effects in stimulus expectation. **Figure 3c** shows the sum of activity across both groups (left hand/right hand), highlighting prestimulus theta activity, that is common to the perceptual classification of non-pain versus pain irrespective of the site of stimulation.

Scale position	Instruction	Score
Leftmost	"No sensation at all"	0
Left to center	"The strongest sensation, that is not yet painful"	49
Center	"Pain threshold - this point cannot be selected"	50
Right to center	"Painful sensation"	51
Rightmost	"Maximum tolerable pain"	100

Table 1: Definition of Rating Scale Anchor Points. Since the middle of the scale cannot be chosen, the ratings can also be dichotomized into a two-alternate-forced-choice (2AFC) dataset between nonpain and pain.

Discussion

Here we used the well theoretically founded QUEST method to efficiently estimate a robust psychophysical threshold between non-pain and pain perception. Using constant stimulation at this threshold enables an analysis of perceptual decisions independent of changes in stimulus magnitude. While we examined threshold intensity at the transition point between innocuous and noxious sensation domains, other points along the pain scale (e.g., 50 on a 100-point pain scale) can also be anchored with the here presented estimation method. In these cases, care has to be taken to account for habituation or sensitization effects across the course of the experiment. Such effects are more likely to occur for higher stimulation intensities.

One critical step in this procedure is to optimally adjust the necessary parameters for the psychophysical function to be fitted by the adaptive procedure. Another important issue is the instruction given to the participant regarding the anchoring of the response scale. The participant should have a clear understanding of where to range in the subjective intensities on the scale. It is thus very important to standardize and repeat these instructions, whenever necessary, to avoid introducing any bias into the ratings. Specifying a scale that is split into a nonpainful and painful side might prove difficult to handle for some participants, since both sensory continua might differ in their respective sensitivity. In this case, when the information from the split-scale is not needed for further analysis, the estimation procedure can also be carried out as a two-alternative forced choice paradigm. Here, the participant just has to decide, whether a stimulus was perceived as painful or not. In case of problems with the rating scale, the estimation will be robust, as long as the participant's response about a stimulus being painful or nonpainful is veridical and false responses are within the limits specified by the delta and gamma parameters.

In cases where the initial thresholding does not converge upon a plausible estimate or rating irregularities become evident, the experiment should be interrupted and restarted. In such cases, it might help to ask the participant about their interpretation of the scale and their subjective perception of the stimulation. If technical errors such as a loose electrode or a faulty connection to the stimulator can be ruled out, it might be helpful to ask the participant about their strategies for dealing with pain. Participants who regularly deal with pain in martial arts or high-performance sports, for example, might exhibit irregular responses despite passing the initial screening. Additionally, social interaction with the participant after the beginning of the experiment and during the breaks should be standardized, as not to induce any effects of experimenter demeanor or induced compliance.

The method outlined here has been demonstrated to be very robust within one experimental session. However, there might be substantial differences in thresholds in the same participants when measured over multiple sessions and days. This might in part be due to circadian changes in pain susceptibility or to changes in arousal or motivation. For heat pain, this has also been shown to be an effect of differing interpretation of the scale range over multiple sessions¹⁸. These problems could be reduced by re-training participants on the scale anchoring in

each session and averaging multiple stimulations into one aggregated rating per trial¹⁹. An additional concern is that reattaching the electrode in a different session might not yield the exact same stimulation intensity and thus may change the estimated threshold.

Using an adaptive estimation procedure such as QUEST, in each iteration, the full set of information from all prior thresholding trials is used to determine the optimal intensity for the next test intensity. This decreases the number of necessary trials while increasing the robustness against inconsistent ratings during thresholding compared to classical methods such as the staircase. The thresholding process could be further optimized by independently gathering data in a pilot experiment to better estimate the slope of the psychophysical function for the desired modality or stimulus type⁷.

Even though the theoretical foundation of the algorithm presented here is sound and we have demonstrated that a robust estimate for exhaustive experiments can be obtained, there are already improved techniques available, that further reduce the number of trials needed to reach robust threshold estimates. These optimized Bayesian methods not only promise less biased results for low trial numbers but also try to fit the position as well as the slope of the psychophysical function in one iteration²⁰.

By using such advanced estimation methods, future research in areas relying on the anchoring of subjective perception can benefit. For one, these algorithms reduce the strain on participants and thus help make the experimental setting more ecologically valid. Additionally, they improve accuracy, not only in threshold experiments, but potentially in all self-report measures suited for psychophysical procedures — a property especially useful for research in the clinical setting.

Disclosures

The authors declare that they have no competing financial interests.

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