Video Article

Measurement of Vibration Detection Threshold and Tactile Spatial Acuity in Human Subjects

Rabih Moshourab[1,2], Henning Frenzel[2], Stefan Lechner[3], Julia Haseleu[2], Valérie Bégay[2], Damir Omerbašić[2], Gary R. Lewin[2]

[1]Department of Anesthesiology and Intensive Care Medicine, Charite Universitätsmedzin, Campus Virchow Klinikum und Campus Charite Mitte

[2]Department of Neuroscience, Molecular Physiology of Somatic Sensation, Max Delbrück Center for Molecular Medicine

[3]Institute of Pharmacology, University of Heidelberg

Correspondence to: Rabih Moshourab at rabih.moshourab@mdc-berlin.de

URL: http://www.jove.com/video/52966
DOI: doi:10.3791/52966

Keywords: Behavior, Issue 115, Vibration detection threshold, forced-choice, psychophysics, tactile acuity, grating orientation, sensory

Date Published: 9/1/2016

Citation: Moshourab, R., Frenzel, H., Lechner, S., Haseleu, J., Bégay, V., Omerbašić, D., Lewin, G.R. Measurement of Vibration Detection Threshold and Tactile Spatial Acuity in Human Subjects. *J. Vis. Exp.* (115), e52966, doi:10.3791/52966 (2016).

Abstract

Tests that allow the precise determination of psychophysical thresholds for vibration and grating orientation provide valuable information about mechanosensory function that are relevant for clinical diagnosis as well as for basic research. Here, we describe two psychophysical tests designed to determine the vibration detection threshold (automated system) and tactile spatial acuity (handheld device). Both procedures implement a two-interval forced-choice and a transformed-rule up and down experimental paradigm. These tests have been used to obtain mechanosensory profiles for individuals from distinct human cohorts such as twins or people with sensorineural deafness.

Video Link

The video component of this article can be found at http://www.jove.com/video/52966/

Introduction

Specialized mechanosensory receptors in the skin mediate the perception of vibration and grating orientation. Each mechanosensory receptor type is tuned to detect distinct features of tactile stimuli[1,2]. This property provides the psychophysical basis for a differentiated assessment of mechanosensory function by using tests that deliver simple sinusoidal oscillations (vibration) or fine gratings. It is known that psychophysical thresholds for vibration perception are lower for high than low frequency vibration[3,4]. Two types of rapidly adapting mechanoreceptors associated with the Meissner and Pacinian corpuscles primarily detect low (10-40 Hz) and high (100-200 Hz) frequency vibration stimuli, respectively[5]. It is thought that the psychophysical thresholds for vibration perception rely to a considerable extent on the activation of these two sets of mechanoreceptors at their best frequencies[6,7]. Tactile spatial acuity is tested by the grating orientation task to determine the finest grating whose orientation can be discriminated by a subject[7-9]. Merkel cell-neurite complex afferents are essential for detecting grating orientation[8,10]. Interestingly, there has been considerable recent progress in our understanding of the molecular basis of how mechanoreceptors detect extremely small tactile stimuli[11]. Mechanosensitive ion channels like Piezo2 and modulators like STOML3 have been directly implicated in the detection of fine tactile stimuli in touch receptors[12-15]. There are already human patients identified with function altering mutations in the Piezo2 gene and it will be important to test whether such patients have touch deficits[16].

The determination of the vibrotactile threshold relies on the delivery of a vibrating stimulus to the skin. Vibration detection thresholds vary with the vibration frequency. Vibration frequency to detection threshold curves were described by von Bekesey, Verillo, and Bolanowski, among many others, from the 1930s up to the 1990s[5,17]. The devices in the early days were based on shakers and power amplifiers and several devices have been commercialized and there are a lot of variation as to choice of stimulator (testing frequency), the diameter of the probe in contact with skin, the use of a surround to limit the stimulation to one area of the skin, and the testing protocol that determines the threshold (see the following references for more detailed insight on the device and testing features)[18-20]. Most devices usually test one site; however, there are new devices that use vertical displacement stimulators equipped with 2 probes to deliver vibratory stimuli with separation ranges that can be varied[21]. Also detection thresholds are measured based on vibration frequency or intensity discrimination with continuous stimulation; or with intermittent vibration stimuli with or without a masking stimulus. Therefore, we recommend that the reader be aware of the plethora of developments in this field.

Here, we describe the components of the device and a step-by-step guide on how to conduct psychophysical tests to estimate vibration detection threshold in human subjects. We then show how to assess tactile acuity manually by using the tactile acuity cube. We use a two-interval, forced-choice paradigm: the stimulus is always presented during one of two intervals and the subject has to indicate which interval has the vibration stimulus. We employ a transformed-rule up and down as the adaptive method that executes a threshold search by changing the stimulus intensity based on the performance during the test. Both psychophysical protocols can be used by investigators as a screening tool for evaluation of alterations in touch sensitivity.

Protocol

The testing protocol was approved by the Charité-Universitätsmedizin Ethics Committee.

1. Vibration Detection Threshold (VDT)

1. Device and Testing Protocol Assembly – Pretesting
    1. Assemble the components of the device according to **Figure 1A**. Place a smooth-surfaced board (40 cm x 80 cm) on a table. Place the brass bar on the board.
    2. Connect the piezoelectric actuator (vibration stimulator) to the controller unit.
    3. Connect the response box and the monitor device to the data acquisition system (see supplemental code file).
    4. Connect the data acquisition system to a computer (or laptop), and to piezo actuator controller unit.
    5. Screw the custom-made stimulating probe to the moving part of the piezoelectric actuator (for specifics of the probe see **Materials**).
    6. Mount the piezoelectric actuator with the probe on the balanced brass bar.

2. Testing Protocol
    1. Script a test protocol that implements the two interval forced-choice and transformed-rule up and down method. See supplemental code file for an outline of the script.
    2. Construct the waveform of the vibration stimulus as a sinusoidal wave and specify stimulus duration, rise and fall characteristics.
        1. Open software (*e.g.*, LabChart). Select Setup > Stimulator.
        2. Choose a custom waveform and configure the stimulator options. Create 2 stimulus waveforms pertaining to each interval (stim1 and stim2).
           NOTE: The waveform stim1 is composed of 3 parts: a delay of 4 sec, followed by a sinusoidal wave of 1.8 sec, and a delay of 1.8 sec (no stimulus). The waveform stim 2 is composed of 3 parts: a delay of 4 sec, followed a delay of 1.8 sec (no stimulus), and by a sinusoidal wave of 1.8 sec.
        3. For the sinusoidal waveform, create new variable parameters for frequency and amplitude. Modify the sinusoidal wave function by inputting the following functions to the rise and fall.
           Rise waveform: $(1-e^{-bt})\cdot$Amplitude$\cdot$sine(frequency), b = 9.1
           Fall waveform: $(e^{-bt})\cdot$Amplitude$\cdot$sine(frequency), b = 9.1
        4. In the data panel of the lab chart, create the set of 35 voltage outputs pertaining to the 35 amplitude intensities (or levels) of the vibration stimulus. See **Table 1**.

    3. Set the starting/default amplitude of the vibration stimulus for the tested vibration frequency in the macro script for the testing procedure (see supplemental code file section).

3. Preparation and Training of Subjects – Testing Session
    1. Inform test subjects about the testing procedure and have them sign a written consent form. To ensure anonymity and fulfill data protection requirements, assign each participant a number.
    2. Seat the subjects comfortably in a quiet room at temperatures between 20-30 °C. Instruct them about the test in a simple and clear manner so that the subject knows what to expect during the testing.
    3. Place the arm of the subject on the board. Pad the little finger with medical dough to minimize movement. Place the brass bar on the board to position the probe on the little finger of the tested hand just below the nail bed. Ensure proper contact between the probe and the skin and adjust the position of the probe to a horizontal position using the water-level. Avoid skin contact with the edges of the circular flat probe.
       NOTE: This ensures that the flat surface of the probe applies around 30 g (0.3 N) to the skin surface. Sharp edges can lead to decreased detection thresholds. The heavy mass of the brass bar prevents the transmission of distracting oscillations from the surroundings to the device and minimizes dissipation of the applied sine wave.
    4. Prior to testing, get the participants familiarized with the setup. Depending on the testing frequency, present both an easy (level 23) and a hard (level 7) to perceive vibration stimulus by varying the amplitude until the subject perceives a vibration to ensure that the experimental procedure is understood.
    5. If necessary, reset the starting (default) amplitude at the selected frequency so that the subject can easily detect the stimulus when the testing protocol starts (see 1.2.3).
    6. Minimize interactions between subject and examiner during the test.
    7. Start the test by running the script that uses the two-alternative forced-choice procedure with the up-down adaptive method[22].
       NOTE: Actions in 1.3.7-1.3.12 are automated by the program script.
    8. In each trial, randomly administer a vibration stimulus during one of the two intervals that are visually indicated to the test subject as "1" and "2" on the screen of the monitor (**Figure 1B**). Have the subject indicate if the first or second sequential intervals contained the vibration stimulus by pressing one of two buttons "1" or "2" on the response box. Let the subject make a guess if he or she is not sure when the stimulus is presented.
       NOTE: The forced-choice technique requires that the subject responds even when the vibration is not perceived.
    9. In a trial series, which consists of 6 to a maximum of 9 single trials, repeat the same vibration stimulus at one amplitude level at least six times consecutively. If the responses are all correct, reduce the stimulus intensity level (down rule) for the subsequent trial series.
    10. Based on the decision rule of the adaptive method (logical operators in script), grant the subject more trials to the same stimulus intensity if the subject makes errors in a trial series. Reduce the stimulus intensity if the stimulus is correctly identified in at least 5 trials, and incorrectly in less than 2 trials.
    11. Increase the stimulus level if the subject makes the 2 incorrect responses in a trial series; or, more than one incorrect response and fewer than 5 correct responses.

12. Document the change in the direction of stimulus intensity, as reversal point. Change the stimulus intensity according to reversal point number: prior to the third reversal point by 4 intensity levels; at the 3rd reversal point by 2 intensity levels; else by 1 level (for more detail see supplemental code file and **Figure 4B**).
13. End the testing when the subject completes a total of 8 reversals.
14. Calculate the VDT by taking the median of the stimulus amplitude value of the last 6 reversals.

2. Tactile Spatial Acuity Test

1. Determine tactile acuity with a two-alternative forced choice grating orientation test using the Tactile Acuity Cube (TAC). The TAC is comprised of 6 sides each containing a grating (bar and groove) whose widths are 0.75 mm, 1.25 mm, 1.75 mm, 3.0 mm, 4.5 mm, and 6.0 mm.
2. Seat subjects in a quiet room at temperatures between 20-30 °C and instruct on the task.
3. During the experiment, blindfold the test subjects using shielded eyeglasses. Place the dominant hand on a table with the palmar surface facing up.
4. On each trial, apply the TAC to the finger pad at one of two grating orientations: vertically (parallel direction) or horizontally (transverse direction) aligned to the long axis of the finger. Randomly choose the order of the grating orientation for each trial.
5. Apply the gratings of the TAC for 2 sec to the finger pad of the index finger so that the cube exerts its whole weight on the finger (233 g). Avoid pressing the TAC on the finger pad.
6. Ask the subjects to determine the orientation of the alignment before the cube is removed from their finger.
7. Avoid movement of the participant's finger because it might provide a cue to the orientation. Discard the trial if the experimenter senses that the finger has moved.
   NOTE: Beware that small finger movements might not be detected by the experimenter in the procedure.
8. Employ a two-down and one-up adaptive method in the staircase algorithm.
9. Start with the largest grating, 6.0 mm.
10. Decrease the grating width after two correct identification of the orientation (correct response).
11. Test the next, smaller width and continue with the stepping rule until the subject makes an incorrect response and document the grating width as a reversal point.
12. Increase the grating width stepwise again until the two orientations of a width are determined correctly again.
13. End the test after completion of thirteen reversals.
14. Calculate the tactile grating orientation threshold by taking the median of the grating widths of the last 10 reversals.

Representative Results

The piezoelectric actuator provides the vibration stimulus to the subject. The vibration stimulus has a total duration of 1.8 sec and is presented only once during a trial during the first or second interval (**Figure 2A**). The rise and fall time at the onset and offset of the stimulus is determined by the functions $(1\text{-}e^{-bt})\cdot$Amplitude$\cdot$sine(frequency), and $(e^{-bt})\cdot$Amplitude$\cdot$sine(frequency), respectively, where b is set at 9.1. The rise and fall time at onset and offset are 500 and 600 msec, respectively, and are independent of the testing frequency and amplitude. The duration of the stimulus between the onset and offset phases is 700 msec. The gradual rise and fall ensures a smooth stimulus delivery.

Vibration detection thresholds are dependent on stimulation frequency because they are mediated by different sensory receptors. According to the human psychophysical tuning curve, thresholds lie between ~20 nm to ~45 μm[5]. Therefore, a set of 35 levels of stimulation (range from 18 nm to 45 μm) of the vibration waveform is constructed (**Figure 2B**) whose amplitude values are arranged logarithmically (base 10; $\text{stimulus}_{n+1} = 10^{0.1}\cdot\text{stimulus}_n$). This range of amplitudes is designed to allow to test at frequencies ranging from 1-250 Hz. The starting stimulus amplitude is usually set above the average vibration detection threshold for a particular vibration frequency test. Previous observations on frequency tuning curves obtained from both psychophysical studies measured average detection threshold around 300 nm for high frequency (>100 Hz) stimuli, and ~3 μm for lower frequencies (<40 Hz)[5,23,24].

The relationship of the driving input voltage to the output displacement of the piezoelectric actuator measured by the strain gauge sensor (SGS) at 10 and 125 Hz are illustrated in **Figures 3A** and **3B**. The relationship is linear (correlation coefficient, $r^2 = 0.9992$) for the set of vibration intensities used (**Figure 3C**). There was a nearly identical relationship between input voltage and output displacement at sinusoidal waveforms of 20 nm amplitude (**Figure 3D**).

In **Figure 4**, a typical testing session where the vibration detection threshold at the right little finger is determined at 125 Hz. The threshold search starts with a stimulus that is above threshold of amplitude level 23 (676 nm displacement). The experimental session consists of multiple series of single trials. A series consists of up to 9 single trials applied at one vibration stimulus level. A decrease in stimulus intensity (level) in general requires at least 6 sequential correct responses (**Figure 4A**). A change in the direction of stimulus intensity marks a reversal point in trial series track and requires at least two incorrect responses. The magnitude of change in stimulus intensity depends on the reversal number. To rapidly search for the threshold, the stimulus intensity is changed in steps of 4 levels when the number of reversals is less than 3, followed by 2 levels at the 3rd reversal (upward direction). Otherwise, the stimulus intensity changes in 1-level steps to finely determine the threshold (**Figure 4B**). The subject's threshold is calculated by converting the median stimulus intensity of the last 6 reversal points; in this case 401 nm. The transformed-rule up and down procedure converges to the threshold value at which 75% of the responses are correct. This was calculated based on the response sequences leading to a down rule using the following formula: $p^6 + 6p^6.(1\text{-}p)+6p^6.(1\text{-}p)^2$, where p is the probability of a correct response.

In the same subject, the tactile cube experiment was done (**Figure 5**). Each trial consists of 2 stimulations. A two-down and one-up rule is employed and the experiment ends after 13 reversal points. The tactile acuity threshold is 1.6 mm is the median value of the grating widths of the last 10 reversal points. The 2-down and 1-up staircase rule converges to a 71% correct threshold[25].

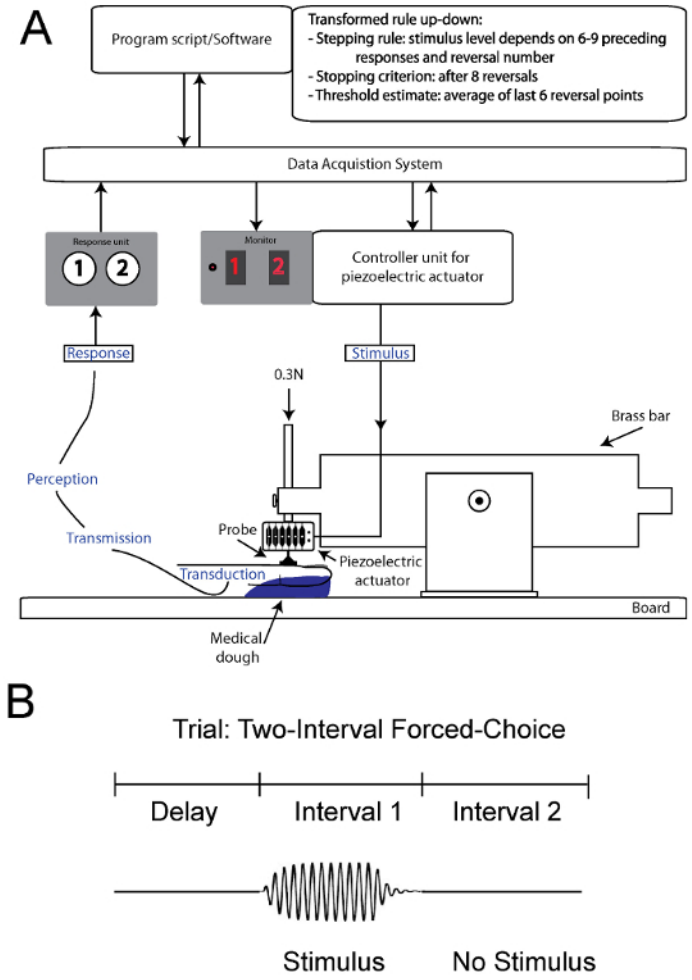

Figure 1: Schematic diagram illustrating the setup components for measurement of vibrotactile thresholds (**A**) and a typical test trial (**B**). Please click here to view a larger version of this figure.

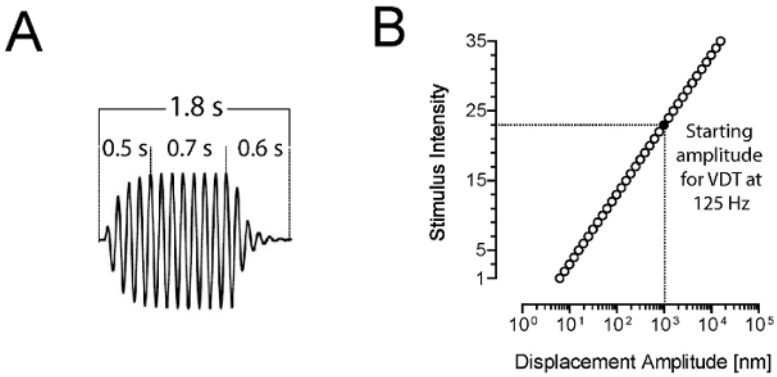

Figure 2: Physical characteristics of the vibration stimulus. (**A**) schematic presentation of the stimulus waveform. (**B**) amplitudes for vibration stimuli (the x-axis has a logarithmic scale). The black circle designates the starting amplitude strength for vibration testing at 125 Hz. Please click here to view a larger version of this figure.

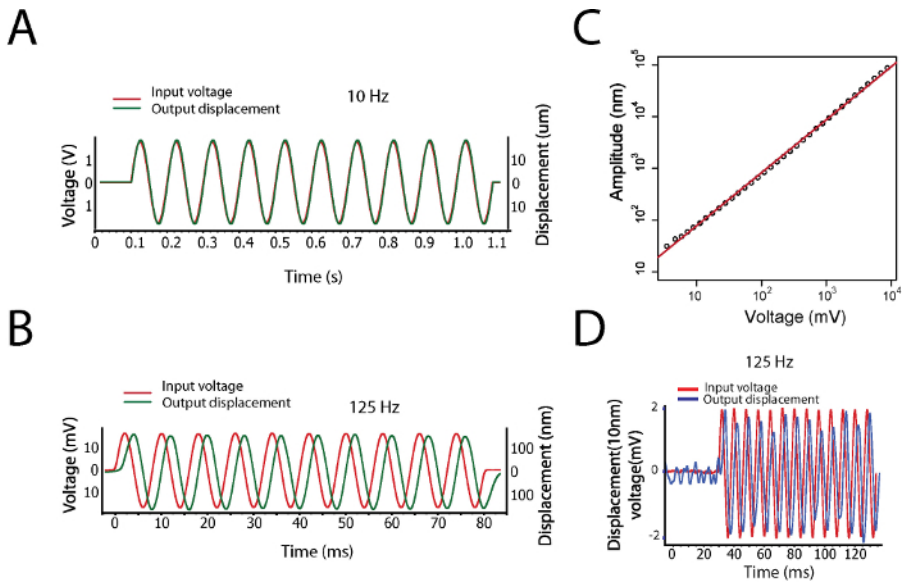

Figure 3: Relationship of the driving input voltage to output displacement of the piezoelectric actuator at 10 Hz and 125Hz sinusoidal input waveform, (**A** and **B**), measured by the SGS integrated sensor. (**C**) the relationship between the 125 Hz driving input voltage and measured output displacements for the set of vibration intensity levels used in testing. (**D**) Nearly identical input voltage to output displacement at the lowest intensity level, 20 nm. Please click here to view a larger version of this figure.

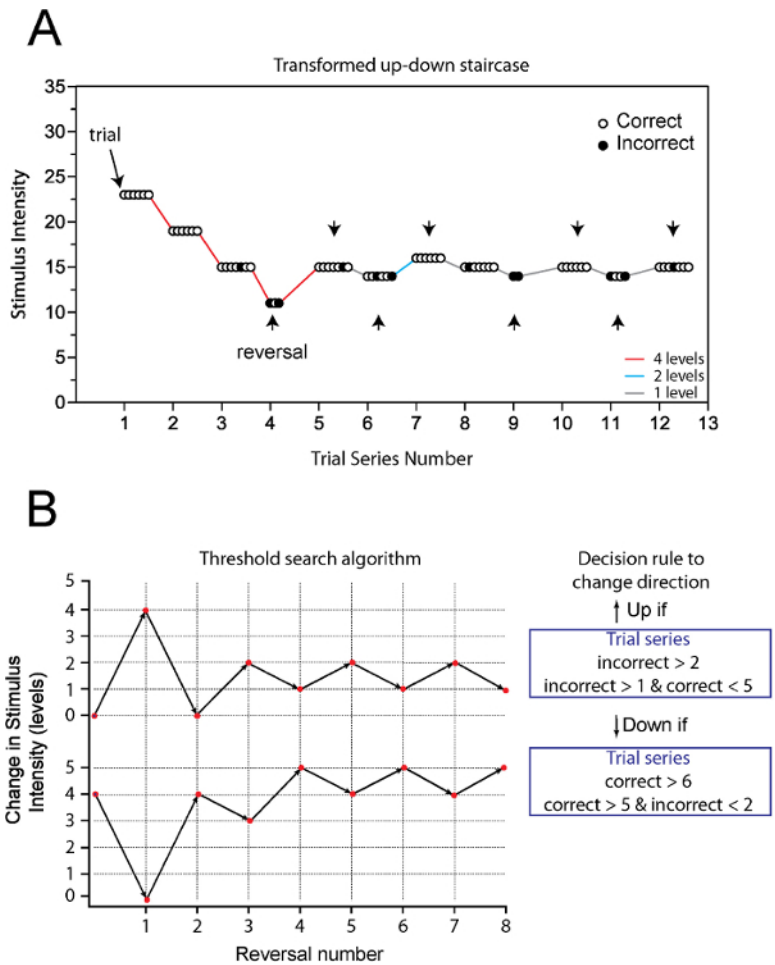

Figure 4: A typical performance of a subject is shown on the two-interval forced-choice vibration detection test. (**A**) the responses of the subject (correct, open circles; incorrect, black circles) are plotted against the trial series number. (**B**) a diagram depicting the adaptive method that searches for the threshold by changing both step sizes and direction (*i.e.*, increasing and decreasing level) across a set of trials. Please click here to view a larger version of this figure.

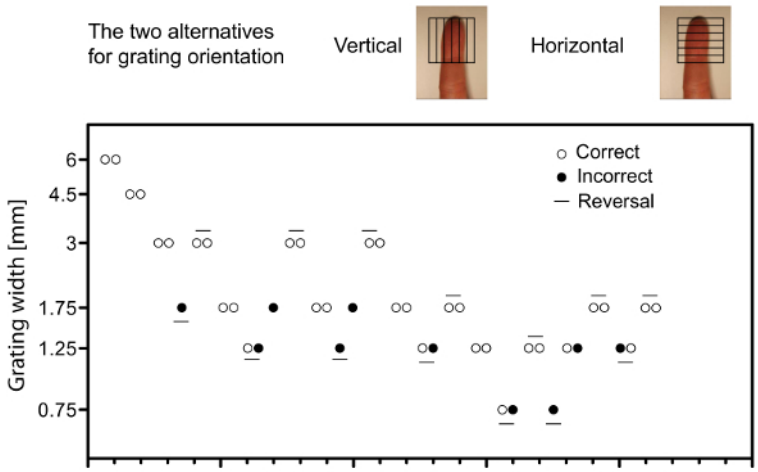

Figure 5: Typical performance of a subject on the two-alternative forced-choice algorithm for the grating orientation task using the tactile acuity cube. The responses of the subject (correct, open circles; incorrect, black circles) are plotted against the trial number. Please click here to view a larger version of this figure.

Stimulus levels and the corresponding output voltages (V) for stimulus amplitudes					
(1 V = 10 µm displacement amplitude)					
1	0.00179	13	0.02839	25	0.45
2	0.00226	14	0.03574	26	0.56652
3	0.00284	15	0.045	27	0.7132
4	0.00357	16	0.05665	28	0.89787
5	0.0045	17	0.07132	29	1.13035
6	0.00567	18	0.08979	30	1.42302
7	0.00713	19	0.11303	31	1.79148
8	0.00898	20	0.1423	32	2.25534
9	0.0113	21	0.17915	33	2.83931
10	0.01423	22	0.22553	34	3.57448
11	0.01791	23	0.28393	35	4.5
12	0.02255	24	0.35745		

Table 1: Stimulus levels and the corresponding output voltages (V) for stimulus amplitudes.

Discussion

The techniques used to evaluate VDT vary as to device specifications, hardware, and testing protocols. The International Organization for Standardization specifies the methods and procedures to analyze and interpret vibrotactile thresholds including recommendations for the various components of a vibrometer (ISO 13091-1 and 2[26,27]) The described testing system abides to the relevant ISO recommendations for testing frequency range (4-125 Hz), method (variant of up-down staircase and forced choice), probe size (smooth edged flat and circular), and testing at room temperature. The setup can be equipped with a sensor determining the probe's position and displacement and optionally can be equipped with a firm surround isolating the vibration stimulus.

The benefits of the described device lie in the following characteristics: availability and easy assembly of the components; adjustable frequency range 1-500 Hz; wide amplitude range (5 nm-90 µm displacement); multiple probe sizes; programmability of the vibration stimulus size and duration; and versatility to psychophysical testing protocol with various adaptive method. Although portable systems are available[28], the setup is transportable and has been used in various settings such as schools, research institutes, and hospitals. Research centers, however, should have a well-equipped workshop to fabricate additional tools and devices needed for the experimental setup. There are commercially assembled and ready for use systems *e.g.* case IV or Medoc, but these systems are configured to deliver only a limited set of stimuli. In contrast, our system can be used to implement a very wide range of stimulation protocols that are only limited by the specifications of the piezo element used. Due to the brass bar size, it is not feasible to test other areas than the fingers and toes. Our device does not use a surround to limit the stimulation to the tested skin area and it is likely that a wider area larger than the actual skin to probe contact is stimulated. Lastly, the procedure requires attention from the subject for a longer period of time, 15 min on average. Usually the first 3 reversals have a larger change in intensity levels (4 levels) and once completed the threshold most likely lies within this range of 4 intensity levels. With progress of the test, fine determination of the threshold is made. There are other stopping criterion which could be implemented and integrated in the adaptive procedure described elsewhere[29].

Several device parameters that can affect vibration detection threshold are adjustable in the setup[17]. These include device parameters such as the contact area of the probe to skin, use of surround encircling the probe to limit the area of stimulation, the selected vibration frequency, wrist posture, and psychophysical testing algorithm. Psychophysical testing algorithms incorporating the method of limits and the staircase method have been used to determine the VDT and there is no consensus favoring a particular method. Vibrotactile thresholds do vary depending on the psychophysical testing method used[20]. Estimates of VDT for 125 Hz are reliably obtained in the range of 50 to 600 nm using the outlined adaptive method in the psychophysical protocol and are in accord with VDT from other studies[5,23]. Furthermore; scripts for different adaptive methods can be easily developed and integrated to execute the psychophysical test for vibration detection threshold[25]. The adaptive procedure we described for the determination of the vibration threshold assures that for each stimulus strength level the subject's correct responses are over 76% correct in order to move down the stimulus strength staircase[22]. Since there are only 2 possible responses, a series of lucky guesses could erroneously alter threshold measurements, especially at low stimulus levels. For this reason we added a modification which is a series of trials at each vibration level in order to minimize such errors. Subjects who show no consistency during trial series – deviations of more than 4 levels between reversal points – are typically excluded from the study. The skin to probe contact and how the probe behaves while stimulating the skin are very important to any psychophysical experiment on touch sensation[30,31]. The piezo actuator is equipped with a compact strain gauge sensor (SGS) has a relatively high bandwidth (up to 3 KHz) and a very good resolution with good repeatability (0.1% of nominal displacement). Therefore, the piezoelectric device has high reliability characteristics especially when it comes to fine indentation even at high static loads. The motion is straight because the piezo (PICMA stack piezo linear actuator) we use is embedded in a guide and this ensures no lateral motion. In addition, the servo-controller can automatically compensate for varying loads or forces.

The assessment of tactile acuity we described here relies on manual delivery of tactile stimuli. The test requires careful application of the stimulus to produce perpendicular deformation of the skin and no shearing distortions that might provide cues to the subject. We choose a slightly different procedure for the determination of tactile spatial acuity and vibrotactile threshold. We did not elect to use a larger step size change initially using the TAC because it is equipped with few levels for grating width (6 levels) and these are not of constant or fixed size but vary from larger to smaller step size change. The change in size of the grating widths between the first 3 levels is 1.5 mm, between the 3rd and 4th level 1.25 mm, and 4th to 6th 0.5 mm. The subject's performance in the grating orientation task is affected by the depth of the indentation caused by the tactile stimulus, the force applied, and finger the size of the finger[32,33]. There are other alternatives to the tactile acuity cube: JVP domes, and the two-point discrimination task. The JVP domes are other alternatives to the tactile cube. The advantages are that the JVP domes have 8 grating widths ranging from 0.35 mm to 3.0 mm. JVP domes can be utilized to assess tactile spatial acuity of the tongue and lips[8], whereas 2 point discrimination task does not rely on identifying grating orientation and is not a valid measure for tactile spatial acuity[34]. Recently, difficulties associated with manual testing have been improved through the introduction of an automated tactile acuity system that applies a forced-choice paradigm to determine tactile threshold for grating orientation[35].

The methods for obtaining the vibrotactile and tactile acuity thresholds we described have been employed to screen individuals for touch-related traits[3,7,9]. In a study conducted by our group, we showed that touch traits are heritable, and that certain genetic lesions causing hearing impairment also influenced touch sensitivity[7]. Moreover, touch sensitivity can be assessed under different experimental condition such as water-induced fingertip wrinkling[9]. It will be important to make quantitative measurements of VDT and tactile acuity in patients with possible function altering mutations in genes involved in regulating mechanoreceptor sensitivity[3,7]. Indeed there has been a recent surge in the identification of genes directly involved in this process like Piezo2 and STOML3[1,13,14,36,37], and rapid progress in this area is sure to identify new genes that regulate touch. The influence of genetic variants in such "touch" genes will be ideally tested in genotyped patients with quantitative psychophysical methods like those described here.

Disclosures

The authors have nothing to disclose.

Acknowledgements

The research and development leading to the above described vibration test was funded by the European Research Council (ERC, ADG 294678) and the German Research Council (grant, SFB665). Thanks go to the Max-Delbrück-Center technical and support staff, Mr. R. Fischer, B. Neumann, and Mr. M. Pflaume, who provided invaluable assistance with the project.

References

1. Poole, K., Herget, R., Lapatsina, L., Ngo, H.-D., & Lewin, G. R. Tuning Piezo ion channels to detect molecular-scale movements relevant for fine touch. *Nat Commun.* **5**, 3520 (2014).
2. Lechner, S. G., & Lewin, G. R. Hairy sensation. *Physiology (Bethesda).* **28** (3), 142-150 (2013).
3. Heidenreich, M., *et al.* KCNQ4 K(+) channels tune mechanoreceptors for normal touch sensation in mouse and man. *Nat. Neurosci.* **15** (1), 138-145 (2012).
4. Mountcastle, V. B., Talbot, W. H., Darian-Smith, I., & Kornhuber, H. H. Neural basis of the sense of flutter-vibration. *Science.* **155** (3762), 597-600 (1967).
5. Bolanowski, S. J., Jr, Gescheider, G. A., Verrillo, R. T., & Checkosky, C. M. Four channels mediate the mechanical aspects of touch. *J. Acoust. Soc. Am.* **84** (5), 1680-1694 (1988).
6. Johansson, R. S., & Vallbo, AA B. Detection of tactile stimuli. Thresholds of afferent units related to psychophysical thresholds in the human hand. *J. Physiol.* **297** (1), 405-422 (1979).
7. Frenzel, H., *et al.* A Genetic Basis for Mechanosensory Traits in Humans. *PLoS Biol.* **10** (5), e1001318 (2012).
8. Van Boven, R. W., & Johnson, K. O. The limit of tactile spatial resolution in humans: grating orientation discrimination at the lip, tongue, and finger. *Neurology.* **44** (12), 2361-2366 (1994).

9. Haseleu, J., Omerbašić, D., Frenzel, H., Gross, M., & Lewin, G. R. Water-induced finger wrinkles do not affect touch acuity or dexterity in handling wet objects. *PLoS ONE.* **9** (1), e84949 (2014).
10. Bensmaia, S. J., Hsiao, S. S., Denchev, P. V., Killebrew, J. H., & Craig, J. C. The tactile perception of stimulus orientation. *Somatosens Mot Res.* **25** (1), 49-59 (2008).
11. Poole, K., Moroni, M., & Lewin, G. R. Sensory mechanotransduction at membrane-matrix interfaces. *Pflugers Arch.* (2014).
12. Schrenk-Siemens, K., *et al.* PIEZO2 is required for mechanotransduction in human stem cell-derived touch receptors. *Nat. Neurosci.* (2014).
13. Woo, S.-H., *et al.* Piezo2 is required for Merkel-cell mechanotransduction. *Nature* **509** (7502), 622-626 (2014).
14. Ranade, S. S., *et al.* Piezo2 is the major transducer of mechanical forces for touch sensation in mice. *Nature* **516** (7529), 121-125 (2014).
15. Wetzel, C., *et al.* A stomatin-domain protein essential for touch sensation in the mouse. *Nature* **445** (7124), 206-209 (2007).
16. McMillin, M. J., *et al.* Mutations in PIEZO2 cause Gordon syndrome, Marden-Walker syndrome, and distal arthrogryposis type 5. *Am. J. Hum. Genet.* **94** (5), 734-744 (2014).
17. Gandhi, M. S., Sesek, R., Tuckett, R., & Bamberg, S. J. M. Progress in vibrotactile threshold evaluation techniques: a review. *J Hand Ther.* **24** (3), 240-255; quiz 256 (2011).
18. Güçlü, B., & Bolanowski, S. J. Vibrotactile thresholds of the Non-Pacinian I channel: I. Methodological issues. *Somatosens Mot Res.* **22** (1-2), 49-56 (2005).
19. Lindsell, C. J., & Griffin, M. J. Normative vibrotactile thresholds measured at five European test centres. *Int Arch Occup Environ Health.* **76** (7), 517-528 (2003).
20. Morioka, M., & Griffin, M. J. Dependence of vibrotactile thresholds on the psychophysical measurement method. *Int Arch Occup Environ Health.* **75** (1-2), 78-84 (2002).
21. Tannan, V., Dennis, R., & Tommerdahl, M. A novel device for delivering two-site vibrotactile stimuli to the skin. *J. Neurosci. Methods.* **147** (2), 75-81 (2005).
22. Zwislocki, J. J., & Relkin, E. M. On a psychophysical transformed-rule up and down method converging on a 75% level of correct responses. *Proc. Natl. Acad. Sci. U.S.A.* **98** (8), 4811-4814 (2001).
23. Gescheider, G. A., Bolanowski, S. J., Pope, J. V., & Verrillo, R. T. A four-channel analysis of the tactile sensitivity of the fingertip: frequency selectivity, spatial summation, and temporal summation. *Somatosens Mot Res.* **19** (2), 114-124 (2002).
24. Kuroki, S., Watanabe, J., & Nishida, S. Contribution of within- and cross-channel information to vibrotactile frequency discrimination. *Brain Res.* **1529**, 46-55 (2013).
25. Levitt, H. Transformed up-down methods in psychoacoustics. *J. Acoust. Soc. Am.* **49** (2), Suppl 2:467+ (1971).
26. International Organization for Standardization. *Mechanical vibration-Vibrotactile perception thresholds for the assessment of nerve dysfunction-Part 1: Methods of measurement at the fingertips.* Geneva, Swizerland: ISO 13091-1 (2001).
27. International Organization for Standardization. *Mechanical vibration-Vibrotactile perception thresholds for the assessment of nerve dysfunction-Part 2: Analysis and interpretation of measurements at the fingertips.* Geneva, Switzerland; ISO 13091-2 (2003).
28. Holden, J. K., Nguyen, R. H., Francisco, E. M., Zhang, Z., Dennis, R. G., & Tommerdahl, M. A novel device for the study of somatosensory information processing. *J Neurosci Methods.* **204** (2), 215-220 (2012).
29. Güçlü, B., & Oztek, C. Tactile sensitivity of children: effects of frequency, masking, and the non-Pacinian I psychophysical channel. *J Exp Child Psychol.* **98** (2), 113-130 (2007).
30. Cohen, J. C., Makous, J. C., & Bolanowski, S. J. Under which conditions do the skin and probe decouple during sinusoidal vibrations? *Exp Brain Res.* **129** (2), 211-217 (1999).
31. Makous, J. C., Gescheider, G. A., & Bolanowski, S. J. The effects of static indentation on vibrotactile threshold. *J. Acoust. Soc. Am.* **99** (5), 3149-3153 (1996).
32. Goldreich, D., & Kanics, I. M. Tactile Acuity is Enhanced in Blindness. *J. Neurosci.* **23** (8), 3439-3445 (2003).
33. Peters, R. M., & Goldreich, D. Tactile Spatial Acuity in Childhood: Effects of Age and Fingertip Size. *PLoS One.* **8** (12) (2013).
34. Tong, J., Mao, O., & Goldreich, D. Two-point orientation discrimination versus the traditional two-point test for tactile spatial acuity assessment. *Front Hum Neurosci.* **7**, 579 (2013).
35. Goldreich, D., Wong, M., Peters, R. M., & Kanics, I. M. *A Tactile Automated Passive-Finger Stimulator (TAPS).* (28) (2009).
36. Coste, B., *et al.* Piezo proteins are pore-forming subunits of mechanically activated channels. *Nature* **483** (7388), 176-181 (2012).
37. Martinez-Salgado, C., *et al. Stomatin and Sensory Neuron Mechanotransduction.* **98** (6), 3802-3808 (2007).

