# SpikerBox—Neural Engineering Workshop
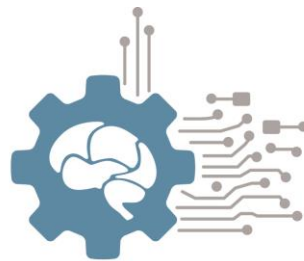
## A Workshop Curriculum for Grades 9-12

Developed as a two-day, two hours/day workshop



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**Workshop Time:** 2- 2 hour sessions or 1- 3 hour session

**Background:** The nervous system operates by taking in natural stimuli (sound, vision, touch, etc.) and responding in the form of electrical action potentials among groups of neurons. The nature of this stimulus-response relationship is dependent on both input into the system (stimuli) and the output of the system (response by a group of neurons), and thus is a defining feature of that particular system. Scientists and clinicians can take advantage of the input-output relationship by recording a neural response and reconstructing the stimulus that likely evoked that response. In the motor system, the activity of a group of neurons is what contracts muscles and allows us to move. If you record from the brain of a paralyzed patient, you would see that their neurons are active just like an able-bodied person even though they can't actually make the intended movement. However, since we know what the input-output relationship is between neural activity and movement, we can use the recorded neurons to artificially create the encoded movement. This is the basis for neural engineering; understanding the relationship between an input and an output so that we may use one to engineer the other.

**Session 1:**

**Objective:** To understand how the nervous system uses action potentials to code responses to sensory stimuli. **Today you will be learning how action potentials encode sensory information by recording from neurons in a cockroach leg in response to sensory stimuli.**

 **Key Words:**

- Input
- Output
- Action potential (AP)
- Electrode
    - Recording
- Encoding
- Decoding

**Key Concepts:**

1. A system can be defined by its input-output relationship
2. Neurons facilitate the inputs and outputs of the nervous system
3. We can directly measure the nervous system by recording from neurons
4. The nervous system encodes information about the world

**Key Questions:**

1. What is the input-output relationship of any system?
2. How do you record from the nervous system?

3. What ways can you stimulate the nervous system?
4. How does the response of the cockroach leg encode information about the stimulus?

**Materials List:** Per group, 2-4 students/group

- Backyard Brains SpikerBox© Bundle, which includes:
  - Recording Electrode
  - Smartphone Data Cable (green cable, Macs and mobile devices)
  - Laptop Data Cable (blue cable, PCs)
  - Stimulation Cable
- Y-splitter
- 2 x audio cables
- Mini audio amplifier/speaker, can be found at RadioShack
- Laptop, tablet, or smartphone
- Audacity, freely available for download for laptops
- Backyard Brains App, freely available for Apple devices
- Cockroach leg or Cricket leg (which you can get at a local pet store)
- Toothpicks
- Tweezers
- Extra sewers pins

## Exercises:  READ THROUGH ALL OF THE INSTRUCTIONS CAREFULLY!

 **A cockroach leg contains sensory spines that are innervated by neurons.** We can record the activity of these neurons and how they respond to stimuli.

**Setting up the SpikerBox**



Figure 1

- Open your kit and remove the SpikerBox from the bottom compartment
- Remove one of the Speakers - turn it on just past the click (don't turn the volume all the way up).
- Take out the black "Y" splitter cable - plug the male (unsplit) end into the Spiker Box (green arrow in Fig 1).
- Take out black audio cable and plug into **INPUT** of speaker- connect speaker to one female end of the splitter cable.
- **MAC:** Remove the **GREEN** cable and plug "SpikerBox" end into the other splitter cable jack, and the "Smartphone" end into the headphone jack of the computer.
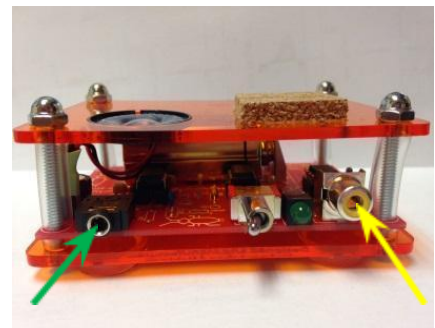
PC: Remove the **BLUE** cable and plug one end into the other splitter cable jack, and the other end into the microphone jack of the computer.

- Carefully pick up the cockroach leg and place it on the cork pad of the SpikerBox. Position it so that the tibia (Fig 2) is hanging off the **RIGHT** edge a little bit. You want to be able to access the tibia for stimulating later.
- Remove the recording electrode from your box and place one pin (lead) into the coxa and the second lead into the femur. Push all the way through so that the leg is secure on the cork.
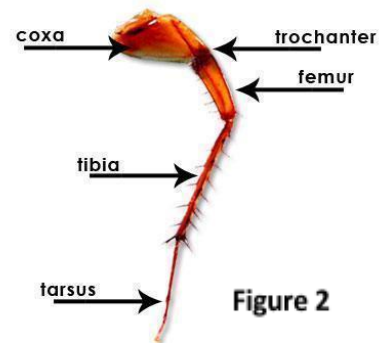
   a. **There are 2 pins that comprise your recording electrode. What is the role of each pin?**

- Plug the electrode into the SpikerBox (Fig. 1 yellow arrow) and turn it on by flipping the switch (a green light should come on and you should hear a noise). You are now recording from a cockroach leg!
- Listen for the little pops in the recording that happen above the general noise. If you're having trouble getting a good recording try repositioning the electrodes ever so slightly. Also to reduce noise and hear the spikes better keep talking to a minimum.

   b. **What are you recording in the leg?**

- Go into Audacity on the computer, it should already be open
- Hit the record button (red circle).
- Go to "View → Fit Vertically" This will allow you to look at your spikes.
- You can zoom in two ways; use both to make the action potentials big enough to easily see.
   o Click on the zero on the y-axis. You may need to click several times. This zooms vertically. (note: if you zoom in too much you can hold shift and then click to zoom back out.

- o Click on the + magnifying glass on the right hand side of the screen. This zooms horizontally, in time, and will make the recording move faster or slower across your screen.
- Once you are consistently seeing good action potentials, hit the "stop" button (yellow square). You will now be able to scroll through your recording.

**Identifying different neurons**

- Find a section of your recording that has a few big action potentials.
- Use the zoom tool (the magnifying glass with no plus or minus toward the left of the screen) to draw a window around the action potentials.
- Pick out action potentials from 2-3 **different** neurons.

**c. How are the action potentials from one neuron different from the action potentials of another? (**Ex. Size, shape**) Why might action potentials differ between neurons?**

**Calculating the firing rate and amplitude of a neuron:**

- Once you have identified a couple of neurons find a section of your recording that has action potentials from each of your neurons.
- Zoom in so that the width of the window (x-axis) is **1 second long!**
- Count the number of action potentials you see **FROM 1 NEURON**! **This is the baseline firing rate!** It is measured in spikes/second or Hertz. You can get a feel for the firing rate just by listening to the recording as well.
- Find the peak and trough of the action potential. Calculate the **amplitude** by subtracting the y-axis values of the peak and the trough.
- Do this for **2-3 DIFFERENT NEURONS**. Find different sections of recording if necessary to help answer the questions in the table below.

**d. Fill in the table for each of your different neurons:** number each neuron for reference.

**When you are done call over an instructor to go over your answers.**

| Neuron | Peak value | Trough value | Amplitude | Baseline Firing Rate (spikes/sec) |
|---|---|---|---|---|
| | | | | |
| | | | | |
| | | | | |

**3. Sensory stimulation –** The neurons in the spine respond to sensory stimulation such as physical bending and air movement.

- Restart the recording of your cockroach leg in Audacity by clicking the record button again. This will start a new recording window.
- Stimulate the spines on the leg by following the instructions below and answering the questions. Work as a group to note where in the recording the stimulation started and ended so that you can zoom in on the section and calculate the firing rate.
- Pick the biggest, or most obvious action potential in your recording to analyze

    a. **Blow on the leg gently**. In general what happened to the firing rate when you tried blowing on it, did it go up, down, no change?

Stop your recording. Select a section of your recording during the blowing that is 1 second long.

    b. **Calculate and record the firing rate. Compared to the baseline firing rate did it increase, decrease, no change?**

    c. **Gently deflect a spine on the leg with your toothpick. Make sure to touch just one spine and not disrupt the whole leg**. What is the firing rate of the neuron now? What happened to the firing rate here: up, down, no change?

     **d. What is the input and what is the output in this system for each type of stimulation?**

e.  **Spine Stimulation** – Work together to fill in the table trying different stimuli. Before performing each experiment make a prediction of what the result will be based on what you have done so far. Write your prediction in the **Prediction** column. Do this as a group.  For the experiments try having one person apply the stimuli, and a second person recording the results in the **Response** column. Measure your response as firing rate in Hertz as you did above. Compare the response firing rate to the baseline you calculated earlier when discussing a **Change** in firing rate.

| Instructions | Stimulus (input) | Prediction | Response (output) Record firing rate (spikes/sec) |
|---|---|---|---|
| Deflect a spine in one direction for a set amount of time, as in Figure A on the right.  Then deflect the spine in the opposite direction (as in Figure B) for the same amount of time. | Direction of spine deflection A.  B.  | A. Will firing rate: (circle one)  CHANGE     NOT CHANGE  B. Will firing rate: (circle one)  CHANGE     NOT CHANGE | A:  B:  If there was a change did it increase or decrease, compared to baseline? |

| | | | |
|---|---|---|---|
| Touch the spine as lightly as you can.<br><br>Deflect the spine as far as you can | Deflect a lot vs. a little | A. Which direction will you deflect the spine and why?<br><br><br><br>B. Which will produce a greater change in firing rate, a small deflection or a large one? | A. Small deflection:<br><br><br>Large deflection:<br><br><br>B. Was your prediction right? |
| Deflect the spine as you have in previous experiments so that you observe a change in firing rate. Hold the spine there for various amounts of time, returning to no bend in between. | Deflect for a long time vs. a short time<br><br>5 sec<br><br>10 sec<br><br>30 sec | Do you expect the change in firing rate to last the duration of the hold?<br><br>5 sec:   YES   NO<br><br>10 sec:  YES   NO<br><br>30 sec:  YES   NO | Record the firing rate in the first and last second of your hold and compare.<br><br>5 sec:<br><br><br><br>10 sec:<br><br><br><br>30 sec: |

**BEFORE MOVING ON TO THE NEXT SECTION WE WILL ALL REGROUP AND DEMONSTRATE STIMULATING WITH MUSIC.**

Up until now you have been stimulating the sensory neurons in your leg by mechanically activating the spines with your toothpick. There are also motor neurons in your leg which, when active, cause the leg to move. You can stimulate these motor neurons by injecting stimulation, in the form of music, into the leg itself.

**What happened when we started playing the song? Did the leg move? Did the leg seem to move to only certain parts of the song?**

Over the next week, think about what in the song is activating the motor neurons and causing the leg to move. Come next week with a list of songs you want to try using as stimulation for the leg. Think about why you chose those songs and what you expect the response in the leg will be. Also think of songs that may not cause leg movement. Here are some ideas to try:

- Songs with a lot of bass vs little bass
- Songs with more speaking (rap) vs more instrumental
- Loud songs vs soft songs

**Homework for next session:**

1. Choose your list of songs

2. Fill in first **3 columns** of the table on the last page.
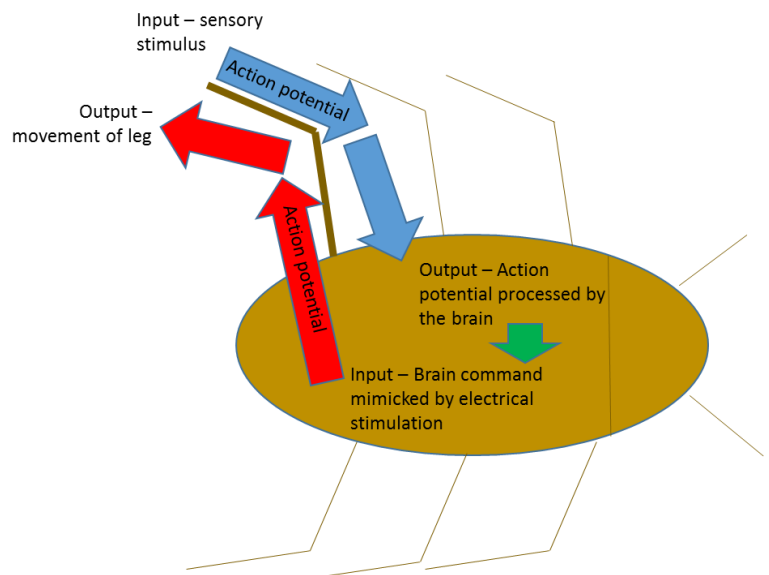
**Session 2:**

**Objective:** To understand how the nervous system uses action potentials to drive motor output. **Today you will be stimulating neurons to produce action potentials which will cause a cockroach leg to move.**

**Review**: Peripheral nerves, those that go to legs, arms, and other extremities, contain both sensory neurons and motor neurons. Last week you manually stimulated the nerve in the cockroach leg. This stimulation elicited action potentials in the sensory neurons which travels to the brain and tells the cockroach something about the external world.  Neurons can also be stimulated by electricity to produce action potentials. Today you will use electrical stimulation in the form of music, sound waves, to elicit action potentials in the motor neurons. Activity in motor neurons causes muscles to contract, resulting in movement. You will explore how different songs activated motor neurons and thus cause the cockroach leg to move. As a review and preview fill in the table below highlighting the difference between sensory and motor neurons.

|  | **Sensory Neuron** | **Motor Neuron** |
|---|---|---|
| **A. What is the input?** |  |  |
| **B. What is the output?** |  |  |

**Key Words:**
- Stimulating electrode
- Encoding
- Decoding
- Sensory neurons
- Motor neurons

**Key Concepts:**
1. Stimulating a sensory neuron vs. stimulating a motor neuron
2. Scientists and clinicians can use this information encoding to simulate neural intent for motor output in injured patients or prosthetics.

**Key Questions:**
1. How is electrical stimulation like/unlike sensory stimulation?
2.  How might you use sensory responses from one cockroach to control the leg movement of a second cockroach?

**Setting up the SpikerBox**

**Figure 1**

- Open your kit and remove the SpikerBox from the bottom compartment
- Carefully pick up the cockroach leg and place it on the cork pad of the SpikerBox. Position it so that the tibia (Fig 2) is hanging off the **RIGHT** edge a little bit. You want to be able to access the tibia with your manipulator for stimulating later.
- Remove the recording electrode from your box and place one pin (lead) into the coxa and the second lead into the femur. Push all the way through so that the leg is secure on the cork.
- Plug the electrode into the SpikerBox (Fig. 1 yellow arrow) and turn it on by flipping the switch (a green light should come on and you should hear a noise). You are now recording from a cockroach leg!
- Use the speaker on the SpikerBox to listen for the little pops in the recording that happen above the general noise. If you're having trouble getting a good recording try repositioning the electrodes ever so slightly. Also to reduce noise and hear the spikes better keep talking to a minimum.
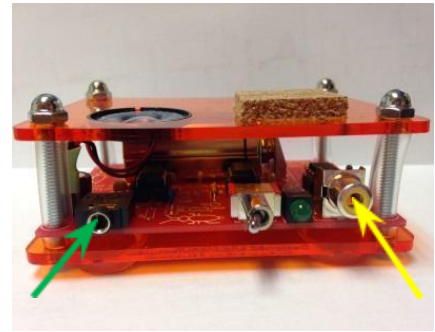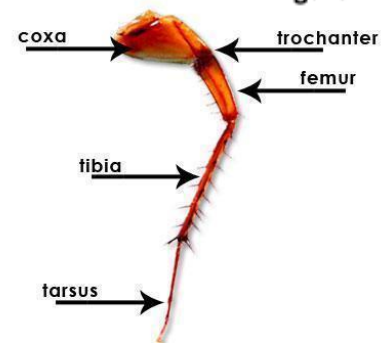
**Figure 2**

coxa  trochanter
femur
tibia
tarsus
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**Setting up electrical stimulation**

- Now that you know you have good electrical contact with the neurons in the leg, it's time to start stimulating electrically!
- Turn your SpikerBox off and unplug the recording electrode from the box, BUT NOT FROM YOUR LEG!
- Plug the Y-splitter into the audio jack of the computer
- Remove the stimulating cable from your kit.
- Attach the red and black clips to each pin in the leg.
- Plug one end of the audio cable into the Y-splitter and the other into the INPUT of the RadioShack speaker.
- Turn the volume on the Radio Shack speaker all the way down (just above the click)
- Turn the volume on the device controlling the music to **HALF WAY**!
- When controlling the volume of the music use the computer or phone, NOT the RadioShack speaker (these two volume sources will multiple together so you need to keep one very low)

What is your general hypothesis based on the songs you have chosen? (Ex. Songs with bass will cause movement while songs without bass will not). When you have your hypothesis show an instructor.

**Hypothesis:**

**Conclusions:**

How would you describe the movements of the leg in reaction to the songs you picked?  Did the leg deflect a lot?  Did it move fast?  Why do you think each song caused the leg to act the way it did?  Was your initial hypothesis correct?

**Song Stimulation**: Cue up your songs and work through the table. Start the volume of the song at half way. If the leg doesn't move turn to ¾ and full until you see movement. This is the threshold volume of the motor neurons you are stimulating for that particular song. If the leg is already moving at ½ volume, turn down to ¼ or until movement stops. After you've tested all of your songs fill in the **Conclusions** below your hypothesis. Think about the results of each of your songs individually as well as a whole how the leg responded to stimulation.

| What song did you pick? | Why did you pick it? | Do you expect the leg to move when you play this song? | Did it move? | Threshold volume | How did it move? To a beat? Randomly? Describe |
|---|---|---|---|---|---|
|  |  | YES      NO | YES      NO | ¼    ½    ¾    FULL |  |
|  |  | YES      NO | YES      NO | ¼    ½    ¾    FULL |  |
|  |  | YES      NO | YES      NO | ¼    ½    ¾    FULL |  |
|  |  | YES      NO | YES      NO | ¼    ½    ¾    FULL |  |