Project Title 'Environmental sensor networks based on plant electrical signalling

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Report Title

Development of a Low Cost Device to Measure Plant Electrical Activity based on a Programmable System on a Chip (PSoC ®)

Summary

There has been much research into the electrical activity of animal cells over the past few decades and how this activity governs their physiological activities. In contrast there has not been nearly as much research into the electrical activity of plants cells, or how this activity can affect their physiology. This has changes in recent years as it has been found that the electrical activity governs many physiological processes in plants and can be used to monitor the health of the plant. While a lot of quality work has been done in the field recently, the sheer volume of plants and different conditions they exist in means that a lot of research needs to be performed to fully develop the potential in the field. One problem with performing research in this area is a lack of cheap equipment to allow more researchers to perform experiments. In this paper describes a low cost open source device that can accurately measure the electrical activity of plants. Because it is open source, other researchers can take the base design and add any features that would be beneficial to their specific purposes and experiments.

Report and outcomes
Materials and Methods:

Hardware:

To develop our device we started with a Programmable System on a Chip 5LP, which has a delta sigma ADC used to collect the voltage activity of the plant, 2 voltage DACs used to calibrate the analog signal chain and to offset any DC component in our measurements and a built in USB 2.0 component to send the data to a computer for analysis. The analog signal chain used in our device is shown in Figure 1 and the schematic at the top of Figure 2. To condition the signal before acquisition we first use an instrumentation amplifier (AD623) to produce a signal that is proportional to the difference between the ground electrode and the electrode used to measure electrical activity. The AD623 is a very versatile single supply instrumentation amplifier that allows for voltages on the input to be below the negative voltage supply rail, which is very useful in measuring plant activity because it allows the device to only need a single power supply and still allow for the measurement of electrical potentials that may be below the ground potential of the device. The AD623 also allows for a reference voltage to be applied to the output signal, this feature allows us to cancel any DC component measured without the need for a high pass filter to remove the DC component. The output of the instrumentation amplifier is then put through a 2-pole low-pass filter Bessel filter (our design uses a multiple feedback topology for the filter). Finally the PSoC 5LP's delta sigma ADC is used to acquire the voltage data. It should be noted that the delta sigma ADC has superior anti-aliasing properties that greatly reduce the amount of anti-aliasing filtering that has to be done to the signal [8, 9].

The instrumentation amplifier can use a resistor to increase the gain of the circuit, but we found out that the amplitude of the potentials on *mimosa pudica* was large enough that this gain was not needed, though it could be used in other iterations of the circuit to measure other plants. The delta sigma ADC that comes with the PSoC 5LP has a differential input with programmable gains settings of 8, 4, 2 and 1. Also it can also have different configurations that can use different reference voltages with the lowest being ±1.024 mV and the lowest being ±0.064 mV. When the ADC is programmed to measure with 12-bit accuracy it can theoretically have a resolution of 2 μV. Because most applications for measuring plant electrical activity will have noise higher than this limit, the PSoC 5LP should be more than accurate enough for any application in this field.

Because the ADC can measure in a differential mode we use a DAC to serve as a reference on the negative input of the ADC and as a reference to offset the output of the instrumentation amplifier. In this configuration the device can measure the electrical activity of a plant from a single power supply such as the voltage from a USB connection or a battery.

To calibrate the signal chain another voltage DAC is used and the output pin of the DAC can be connected to the input of the instrumentation amplifier. Then the DAC gives a series of voltage steps to 500 mV, which is measured by the voltage measuring circuit and any offset or gain errors in the circuit can be measured and compensated for.

The sampling rate of the ADC is set by a 16-bit timer than fires an interrupt service routine (isr) that takes the current value of the ADC and puts it in the USB-UART. The USBUART is a component in the PSoC that emulates a COM port on the computer side and allows high-level communication functions on the PSoC device side [26].

<u>Software:</u>

To control the device a graphical user interface (GUI) was written in the Python Software Language with the Tkinter library. The GUI allows the user to start and stop recording from the device, displays the data, and allows the user to save the data to a file. The GUI also allows the user to set the sampling rate and the reference voltage level. Figure 3 shows a screenshot of the GUI.

Results:

A picture of our device is shown in Figure 4 and an experimental setup shown in Figure 5. In this setup we are measuring the action potentials of *Mimosa pudica*. A typical result of an action potential reading is shown in Figure 6. In Figure 6 the blue line is the voltage readings from our device. The green line in Figure 6 is a line fitting the data with a single activating and a single inactiviation exponential as shown in Equation 1.

$$y(t) = A_{act} \cdot e^{-t/\tau 1} - A_{inact} \cdot e^{-t/\tau 2}$$

Equation 1

The red line overlaying the data in Figure 6 is a fit to 2 activating and 2 inactivating exponentials as shown in Equation 2.

$$y(t) = A_{act1} \cdot e^{-t/\tau 1} + A_{act2} \cdot e^{-t/\tau 2} - A_{inact1} \cdot e^{-t/\tau 3} - A_{inact2} \cdot e^{-t/\tau 4}$$

Equation 2

As can be seen from the different fitted lines, the action potentials of *Mimosa pudica* are fit to 2 activating, and 2 inactivating exponentials much better than they are to single exponentials. To more clearly see the 2 activiating exponentials, Figure 7 is a close up of the rising phase of the action potential. It should be noted here that these are extracellular recordings so a decreasing extracellular potential is the same as an increasing intracellular potential. From Figure 7, it can clearly been seen there is a fast (~10 msec) and a slow (~300 msec) component. Our interpretation of this that the fast component is a set of mechanical sensing channels that initiates the action potential and the slower component is a ion channels that get activated in response to the opening of the mechanical sensors. There are also 2 inactivation exponential time constants, again suggesting there are 2 different ion channels responsible for the action potential in *Mimosa pudica*.

Discussion:

We were able to develop a device that could accurately record the action potential of *Mimosa pudica*. In the future we will also test the device by measuring the action potential the Venus flytrap. With the large amount of analog capabilities of the PSoC 5LP, our future plans for the device also include adding a second voltage sensing electrode so that action potential speeds can be measured, and also a stimulating electrode that can apply voltage or current to the plant to stimulate it.

The action potential of the *Mimosa pudica* is composed of 2 activating and 2 inactivating exponentials. Given the large different in time constants for the 2 activating phases, we assume this is due to 2 different ion channels, a fast acting ion channels which is most likely a mechanosensing channel, and another ion channel that propagates the initial signal after it reaches threshold.

Conclusions:

Our device can operate as a low cost open source platform for measuring plant electrical activity. While our platform can serve as a base, there are still a lot of analog functional units on the PSoC 5LP that could be used to make other application specific devices to other plant measurements.

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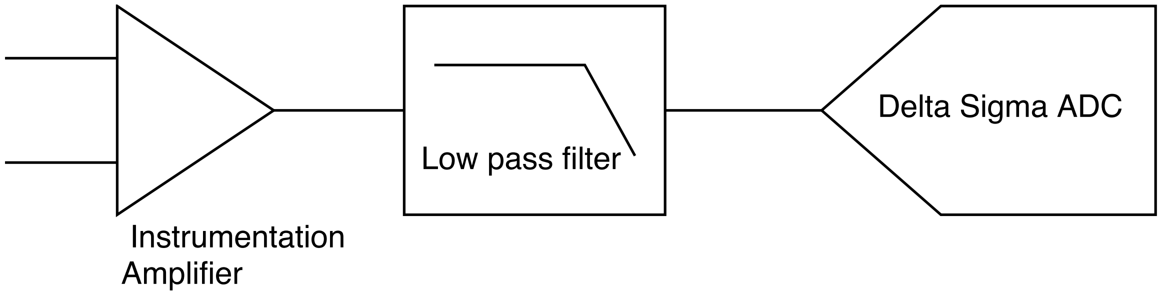

Figure 1: Circuit block diagram of the signal chain for the voltage sensing circuit

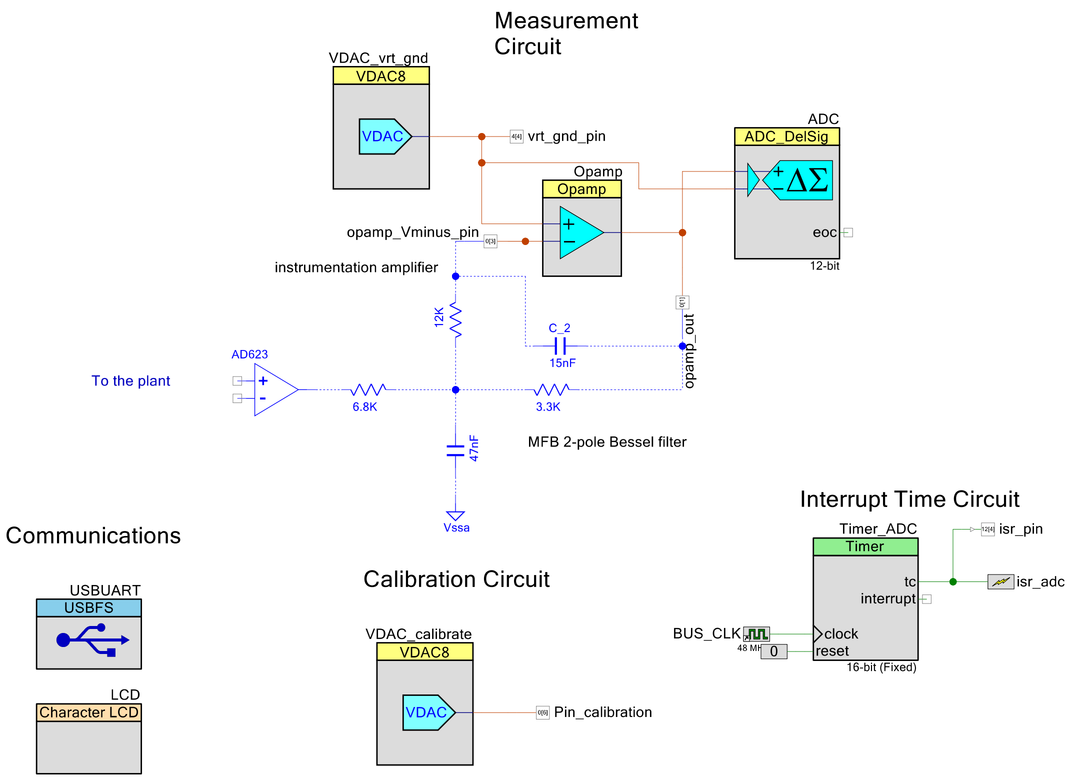

Figure 2: Topdesign schematic for our plant measuring device

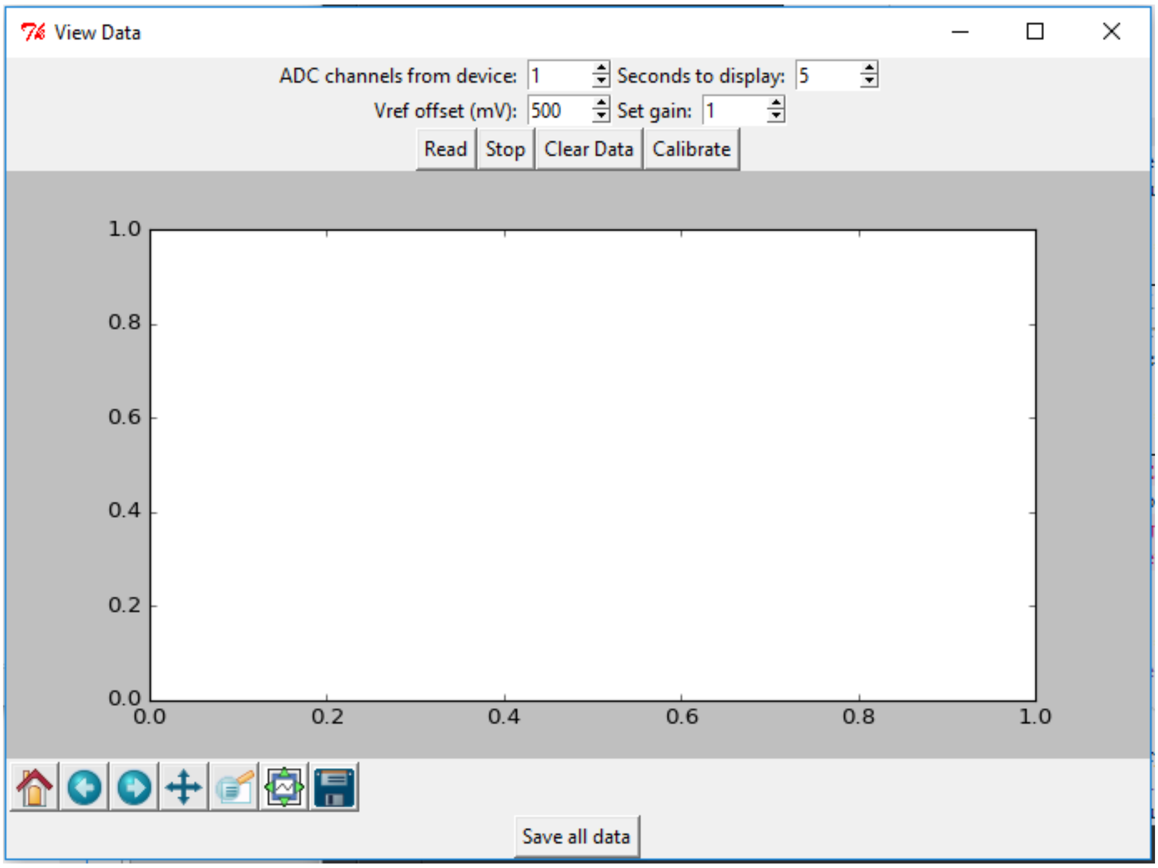

Figure 3: Graphical User Interface for our plant device

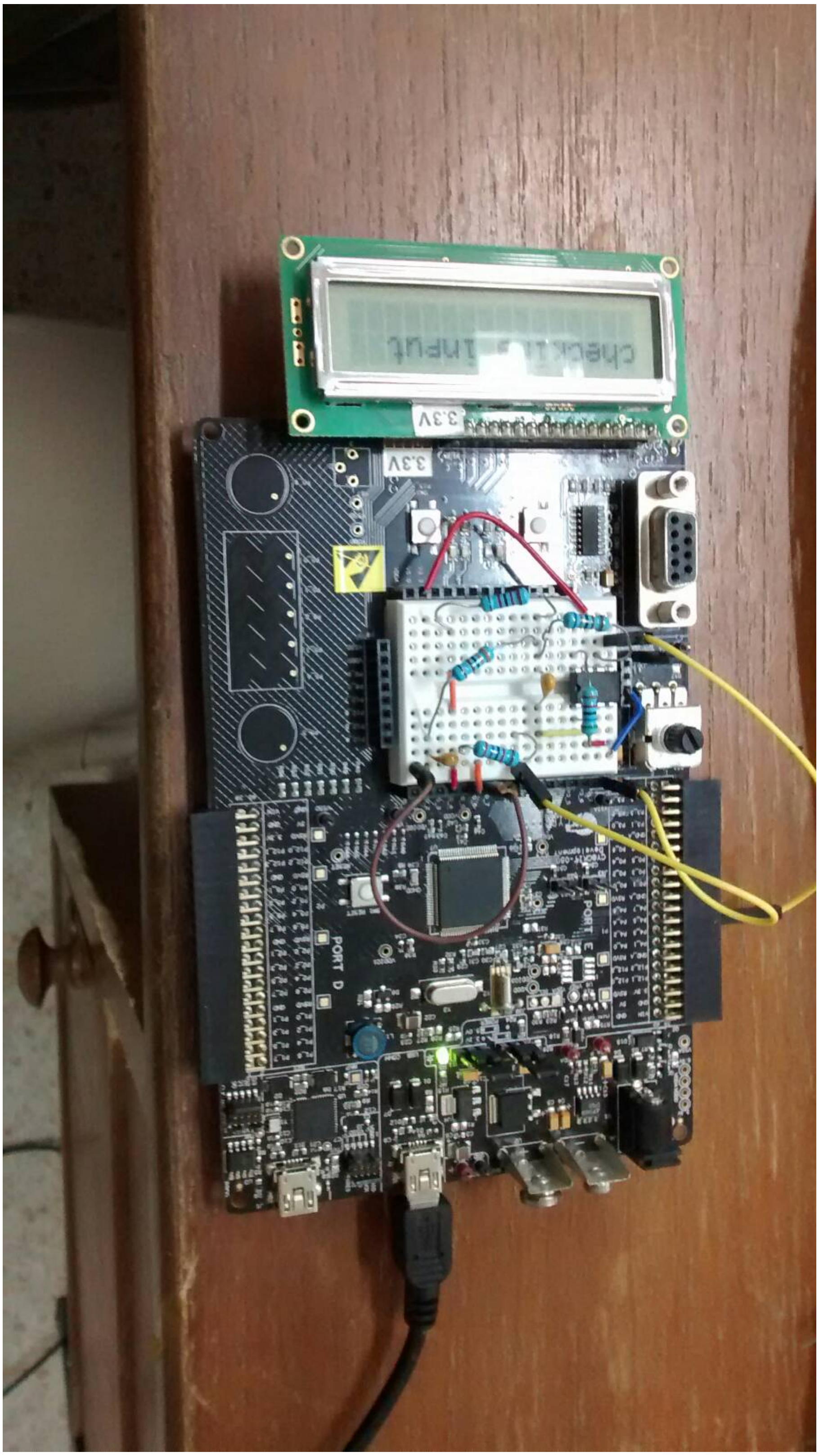

Figure 4: Picture of our plant electrical measuring device

Figure 5: Measuring plant electrical activity setup

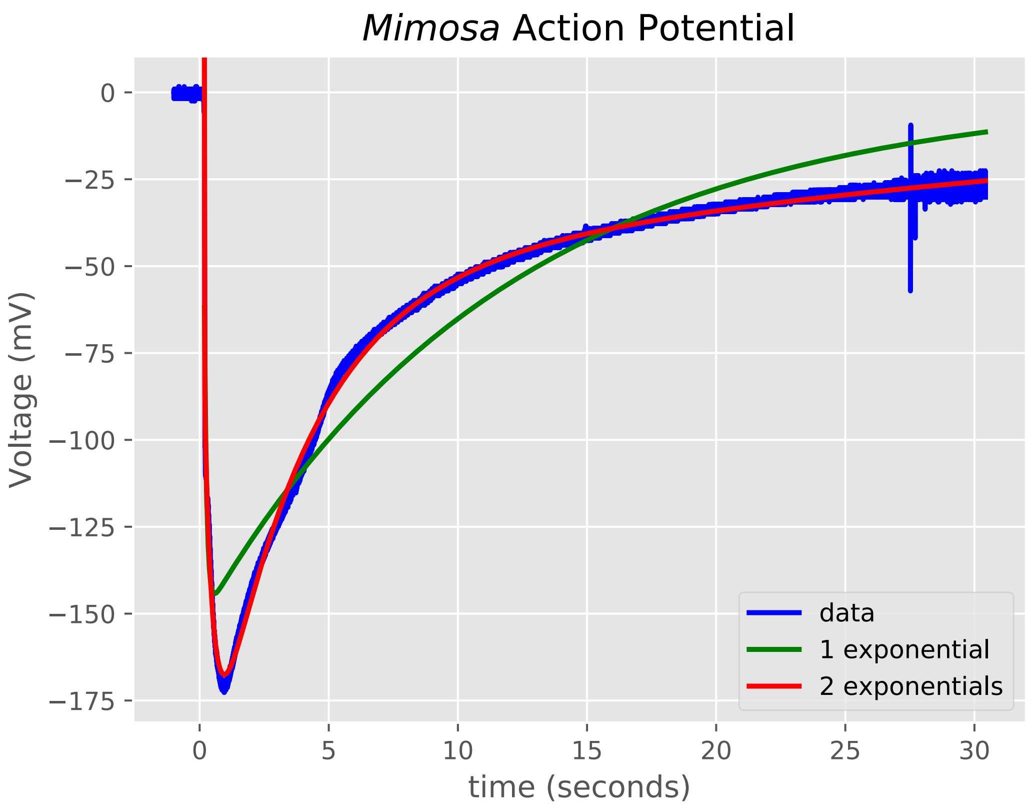

Figure 6: Action potential of *Mimosa pudica*

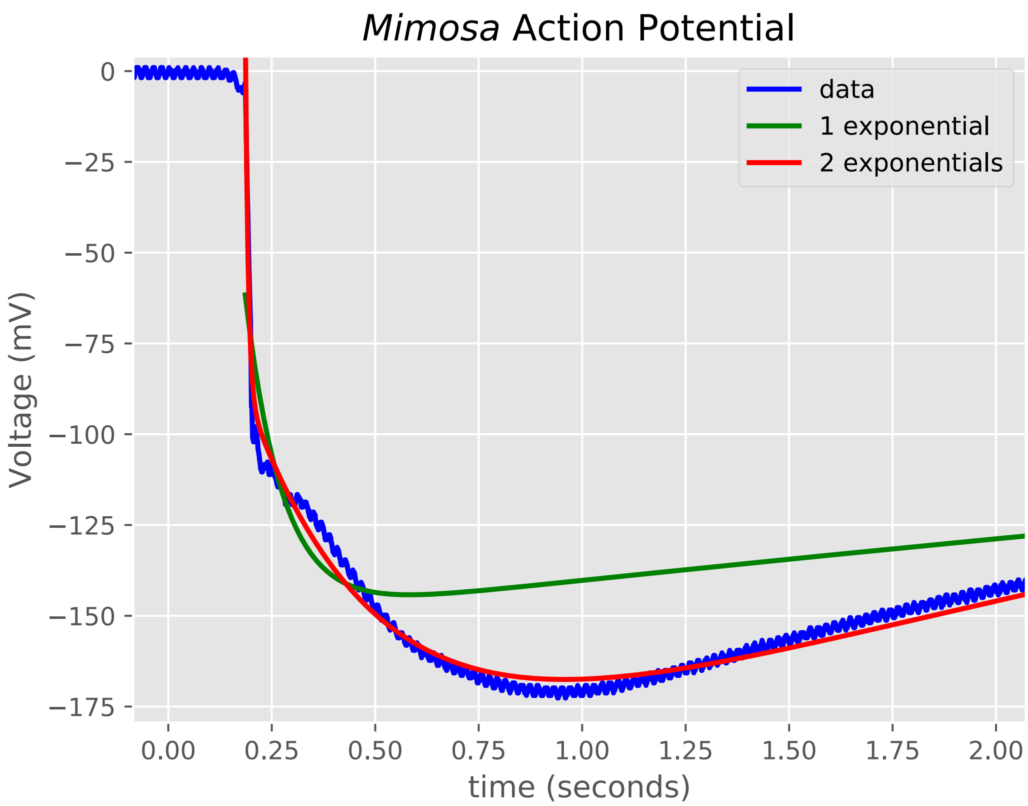

Figure 7: Close up of rising phase of *Mimosa pudica* action potential

Expenditure

The summary of the budget can be found in Openplant report Supplementary file 1.

Follow on Plans

The equipment ordered included development kits (from Cypress Semiconductor) to design and test our plant electrical activity prototypes. We ordered a few development kits to test which Programmable System on a Chip (PSoC) performed the best. We got PSoC 5LP development kits to test as they have very powerful analog components and a high speed ARM Cortex-M3 processor to make a device that is able to measure at high speeds. To test if a portable field device could be used we also got PSoC 4200M kits that have less analog components and a slower ARM Cortex-M0 processor, but it is designed to use lower power so it can be used remotely to sense plant electrical activity. We also ordered raspberry pi to communicate with our device and could display the plant's electrical activity to a user. A few wireless options were also tested to see if the device could work wirelessly so that the device could be deployed in the field. We also ordered a soldering kit to put the device together and a USB based analog / digital oscilloscope to debug our device.