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Reading Electrical Potentials in Vitisvinifera, using Signal pre-processing and Measuring their Response to Heat Shocks

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Abstract—*In an era where Electroencephalography (EEG) and Electrocardiography (ECG) are redefining the field of biophysics, opening up the possibility of Brain or Heart Computer Interfaces, and Electromyography (EMG) is redefining prosthetics among other fields, offering insight into the internal health and structure of the human body, very little research of the same type has been conducted with plants, especially in India. In the case of acute pulmonary embolism in humans, for example, documented researches have offered insight into the changes in ECG data during the condition(JT. 2011), offering a new mechanism to diagnose the case, provided vision into the internal status of a living being using electrical signaling. Such research has not been carried in the field of plant pathology, since electrodes for the same purpose are difficult to create and data acquisition systems for experimentation are not easily available in India. Since plants do not have a formalized nervous system as do human beings, the presence of electrical potential is rarely recognized by farmers in India. One of the aims of this experiment was promoting this recognition, by creating the electrodes required to perform data analysis and measuring these signals using a Raspberry Pi- a relatively easy to acquire system for data. A simple Signal Processing algorithm was then developed to refine the digital signal through MATLAB R2017a (Version 9.2) Processing Environment, and test whether temperature had any impact on the values of the signals acquired, as an initial research. Hence after application of Savitsky-Golay Derivatives and Multiplicative Scatter Correction, statistical means of each of the signals were calculated over the sample sets, and the differences observed between 2 plants kept in close proximity were observed, hereby labeled plant 1 and plant 2. These differences were then compared using a statistical equivalence test- the two one-sided test (TOST). Usage of spectrogram function in MATLAB, which computes the Short Term Fourier Transform was attempted, but could not yield appropriate values due to the low sampling rate of the ADC. The study was conducted to check whether changes in the electrical signals could become representatives of heat shocks at different times of the day. The standard equipment to measure electrical signals in plants is very expensive, and one of the objectives of the research was to check whether any large or distinct changes could be identified using a lesser expensive setup, featuring a Raspberry Pi. The rationale behind the experiment was to promote the recognition of electrical signals in plants in India, and encourage greater study into them in the field of plant pathology and agronomy to leverage their possible use as biosensors in agriculture.*

Keywords*: Electrical Potential (EP), Action Potential (AP), Variation Potential (VP), Heat shock, Two One- Sided Tests (TOST), Multiplicative Scatter Correction (MSC) ,SavitskyGolay Derivatives*

Biological Background, and existing research

Burdon- Sanderson first discovered electrical activity within plants in 1873(Burdon-Sanderson 1873), and these signals have since been recognized by biologists as being the primary method of communication in plants, perhaps of more significance than chemical signaling through hormones or other mechanisms. It has been established that in certain plants, these signals are representative of physiological activity, and often originate in the roots travelling through the vascular system(Fromm and Eschrich, Electric signals released from roots of willow (Salix viminalis L.) change transpiration and photosynthesis 1993)(A. Volkov 2000)(Volkov, et al. 2004).

These signals have been recognized in lower plants- algae such as *Characeae*- as well as more developed ones- such as *Mimosa* and *Cucurbitaceae.* However, until recently, very little research until very recently (Gil, et al. 2014)had gone into fruit crops or more specifically woody perennial plants such as Vitisvinifera, especially in a developing country like India, where grape was one of the largest agricultural exports, and an economically significant crop.

Electrical potentials (EP) are of three major categories within plants: Local Electric Potentials (LEP), Variation Potential (VP) and Action Potential (AP) (Xiaofei, et al. 2009). LEP is only a local response and is not transmitted, although it is induced by sudden changes in immediate environments. Of more significance are AP and VP, which are transmitted along the plant. Action potentials transmit at uniform velocities and amplitudes and propagate through the plasmodesmata (Stahlberg, Cleland and Van Volkenburgh, Slow wave potentials–a prop- agating electrical signal unique to higher plants 2006) (Davies 2004) (Fromm and

Lautner, Characteristics and functions of phloem-transmitted electrical signals in higher plants 2006), and are either present or not. Variation potential, on the other hand, have a changing amplitude and intensity according to the stimulus, and propagate hydraulically(Davies 2004)(Stahlberg, Cleland and Van Volkenburgh, Slow wave potentials–a prop- agating electrical signal unique to higher plants 2006). The transmission mechanisms are not fully understood, but a general distinction is made that action potentials are caused by non- jeopardous stimuli, while variation potentials might have correlation with stimuli that can be potentially dangerous to plants. Hence AP provide less knowledge about the stress that causes them, as compared to signals like the Variation Potential, that may provide early insight of stresses on the plant. It is generally regarded that these electrical signals are important for communication inside plants, and like chemical signals, can change in response to a multitude of biotic and abiotic factors like salinity of soil, temperature and soil moisture(Fromm and Lautner, Characteristics and functions of phloem-transmitted electrical signals in higher plants 2006).

Experimental Setup

Figure 1

Figure 2 Electrode on plant bark

Figure 3 Reference Electrode

One year-old grafted Vitisviniferaplants have been used for the purpose, with complete root growth for consistency of result. The grapevine plants have been grown in compost soil as a control, and have been watered (1L) once a day 7 days prior to the collection of experiment data. The plants are placed in Faraday cages (Figure 1) made at home using cardboard cartons and aluminum foil, with equally spaced slits for insertion of electrodes, and for light to pass through. The Faraday cages are used as a mechanism to prevent electromagnetic interference, or EMP shocks to prevent distorting signals that are in the order of millivolts. The Faraday cages were grounded to the soil using copper wires. In both plants, one electrode was placed inside the potting medium (Figure 3), while the other was inserted approximately 8 cm above potting medium (Figure 2). This was important, as EP was likely to be largely composite of VP, based on the position of the electrodes- xylem (Hellman n.d.)- and the type of readings acquired. Two plants of relative equal height (33'' and 34'') were used for testing, and were placed inside a room with large glass walls and access to sufficient natural light. The experiment lasted 10 days with each plant being tested for electric potential three times a day: morning readings, at 0900 hours, afternoon readings at 1500 hours and night readings at 2100 hours. Each set of readings was 100 samples of EP taken at 860 samples per second, with 20 sets of such data. For every time based reading: two reading sets were taken, one at regular room conditions, and the other as the result of a heat shock as temperature increased (Heat shocks were provided using a heating coil from a hair dryer) from its current value to for the time period of the sampling: the hairdryer was rated at 1000 W, and the values of each set of 100 sample data was observed, progressively recorded at higher temperature. During this time, room temperature value fluctuated from 24 degrees Celsius to 32 degrees Celsius. This was done to check possible effects of heat shocks on values of signals.

Sensor Development

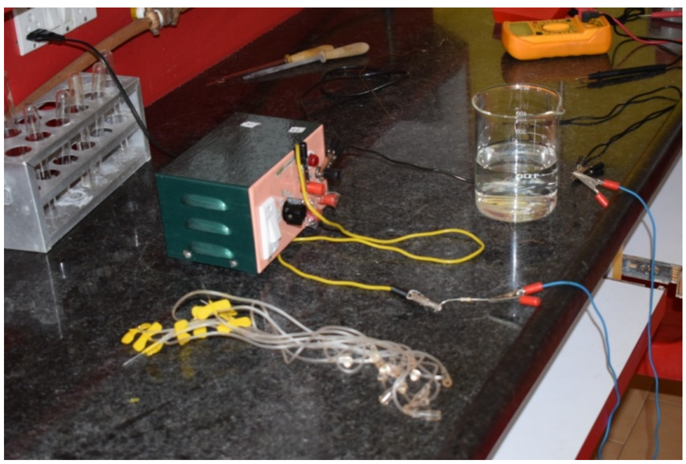

Figure 4: Electrochemistry setup for making electrodes

Electrodes were constructed to insert into the xylem close to the cambium, and previous designs for fruit crop species were utilized (Gil, et al. 2009)(Gurovich and Hermosilla 2009). Electrodes were designed so that they were of the high impedance, yet available locally, for the most accurate reading of voltage- based signals. Ag/AgCl Electrodes were then constructed using 0.35 mm diameter silver wire of 99 percent purity silver. A silver wire of length 7 cm was used in the electrochemistry. A regulated 3.3 V supply was required, with copper wires serving as the anode and the cathode. The positive copper wire was wrapped around the thin piece of silver, and immersed in a chlorinated Hydrochloric Acid solution of 0.1 N, leaving around 2 cm of silver outside. The electrochemistry lasted for around one minute (Figure 4), by which time the color of the silver had darkened to a brown-purple. This was repeated for 10 silver wires pieces, and the coated silver wires were left to dry. A scalp vein needle of diameter 0.7 mm was used next, and the coated silver wires were inserted into the needle from its hole, uncoated end first. Once inserted, the front end of the electrodes was sealed using metal epoxy adhesive, inserted and withdrawing the tip of the needle with utmost precision, so that the glue and the tip form a homogenous surface. 1 M Potassium Chloride was inserted from the back using a standard hypodermic syringe, with emphasis on preventing the formation of air bubbles in the plastic tube behind the needle. This was done by holding the needle in place using a clamp and stand, and pulling the back end vertical so that the solution could flow smoothly. If the solution was dripping from the front end the adhesion process was repeated until the needle tip was sealed. Then the plastic tube was cut until a length that left only the uncoated part of the wire exposed. The electrode in the making was checked for whether it had sufficient KCL, and if it did not, a hypodermic syringe was used to insert it once again. In the final stage, a silicon sealant was used to seal the back end of the cut tube, with the uncoated wire sticking out for voltage measurement. Once the procedure was completed, the electrodes were stored in a cool dry place overnight for appropriate hardening of the sealant.

A standard LM35 temperature sensor was used to monitor the temperature inside the Faraday Cages: it had a linear voltage – temperature response and was calibrated accordingly.

Data Acquisition

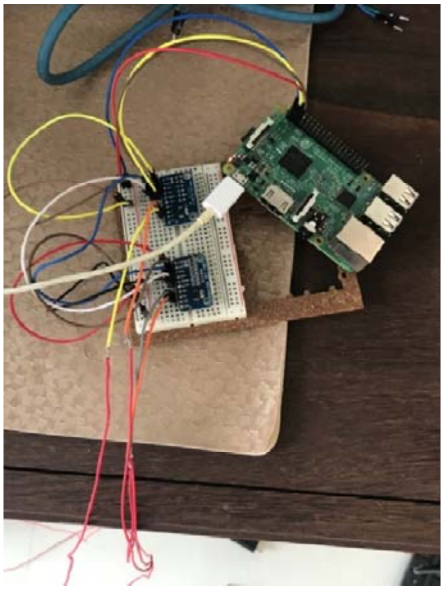

Figure 5: The Raspberry Pi 3 and the ADS1115 ADC

Data is acquired through a Raspberry Pi 3 Model B and Arduino UNO and communicated to the computer through MATLAB interfacing, which is also used as the processing environment. The I2C bus on the Raspberry Pi was used to communicate with an Analog to Digital Converter ADS1115 in both 'continuous' and 'single' reading mode (Figure 5). The ADC contained an internal 4 channel MUX (channel selector) that helped take both differential and single value readings. The electrodes were connected to 860 samples per second ADC, as a data acquisition system or ADC of higher samples per second value was not available. The Arduino was used for the measurement of temperature through the LM35 sensor, as reading multiple channels on a single ADC for various values led to incorrect data sets due to issues in multiplexing. MATLAB R2017a was used for processing of data, and plotting of graphs and spectrograms. As sampling rate was constant, number of samples could be estimated as values taken at increasing values of time.

Signal Preprocessing and Processing

Data was pre-processed to improve Signal to Noise Ratio (SNR) and smoothen the signal using two main algorithms: Savitsky Golay Derivatives and Multiplicative Scatter Correction (Varmuza and Filzmoser 2009), also usually used for NIR Spectral preprocessing.

1) Multiplicative Scatter Correction

In the context, MSC is performed using a subset of the electrical signal data as the calibration set. For each one of these readings, mean was calculated across each of the specified columns of data, and this mean was linearly correlated with the calibrated set values. Then for each of the data sets, using the correlation coefficients obtained, data was corrected to improve signal to noise ratio.

The MSC algorithm for the non- increasing heat data is included in Figure 6.

```
function [xmsc,lambda]=msc(x,fvar,lvar)
if nargin==1
   fvar=input('The first correction sample: ');
   lvar=input('The last  correction sample: ');
end

[m,n]=size(x);
lambda=mean(x);

for i=1:m                                                % each signal set
  l=polyfit(lambda(fvar:lvar),x(i,fvar:lvar),1);         % linear least square fit
  xmsc(i,:)=(x(i,:)-l(2)*ones(1,n))./(l(1)*ones(1,n));   % signals are corrected
end


end
```

Figure 6 Algorithm for multiplicative scatter correction

2) SavitskyGolay Derivatives

A Savitsky-Golay filter was used in the process of smoothening the signal and was particularly important since the data acquisition mechanism did not have a very high rating for samples per second. The ‘sgolayfilt’ function in MATLAB was used. Smoothening is achieved through convolution, in which subsets of close data points are successively fitted with a polynomial of lower degree. A LMS (least mean squares) algorithm forms the basis of the process. Convolution coefficients are derived via a standard set of normal equations that are used to minimize the cost function. For the data set, a first order Savitsky- Golay filter was applied. Since the operation operated on columns, and electrical signal values for a sample were stored in rows, a transpose was taken before this filtering operation.

Once the data was preprocessed, the following process was applied:

I. Data from every 100 sample reading set was recorded and a mean value for each EP value was obtained

II. The mean value across the column of the obtained mean values in I. was calculated to get the overall mean value of the sample for easy representation

III. The samples for each morning, afternoon and evening data were obtained, separately for plant 1, and plant 2 on each of the 10 days in question

IV. Equivalence testing for all six of the data sets- vectors of length 10- was carried out using Two- One Side Sample Tests (TOST). As samples taken were under 15, a *t distribution* must be used to decide whether or not the values contained within the vector are equivalent. For this purpose, the ‘tcdf’ distribution function in MATLAB is used with 18 degrees of freedom. Upper and lower values for difference are set and probability that the difference is lower than the lower limit or higher than the upper limit is calculated. The lower and higher limits are taken as [-0.0005,+0.0005], [-0.0010, +0.0010], and so on in positive and negative multiples of 0.0005 before a probability greater than 0.8 is obtained for the difference in values to be within the interval.

The MATLAB algorithm for the purpose can be seen in Figure 7.

```
function [p1, p2] = TOST(plant1, plant2, d1, d2)

me1 = mean(plant1); %mean of plant 1 differences
me2 = mean(plant2); %mean of plant 2 differences

l1 = length(plant1); l2 = length(plant2); %distribution sample sizes
s1 = std(plant1); s2 = std(plant2); %standard deviations of the distributions

EM = ( ( ( (l1-1).*s1^2+(l2-1).*s2^2 )./(l1+l2-2) ).* (1/l1 + 1/l2)  ).^(1/2);

t1 = ((me1-me2)-d1)/EM;
t2 = ((me1-me2)-d2)/EM;

p1 = 1-tcdf(t1,l1+l2-2);
p2 = tcdf(t2,l1+l2-2);
```

Figure 7Algorithm for TOST

Results and Discussion

Readings were taken over 10 days and general patterns in the same were observed.

During the process of reading data and filtering readings, data was stored according to a code. For example O01221 implied that the data was taken on the first of October (first 3 digits), in the afternoon (4th digit: 1- Morning, 2- Afternoon and 3- Evening), on plant 1 (5th digit: Number of plant) with heat (6th digit: 1- heat shock, 2- room temperature).

An exemplar graph for a set of readings against nth sample has been shown below (Figure 8). The graph represents the first sample for October 2 morning data at room temperature for Plant 1. The same sample, with Savitsky- GolayDerivatives and Multiplicative Scatter correction applied has also been included below (Figure 9).As can be seen the signal has been smoothened, eliminating any random scatter inside it.

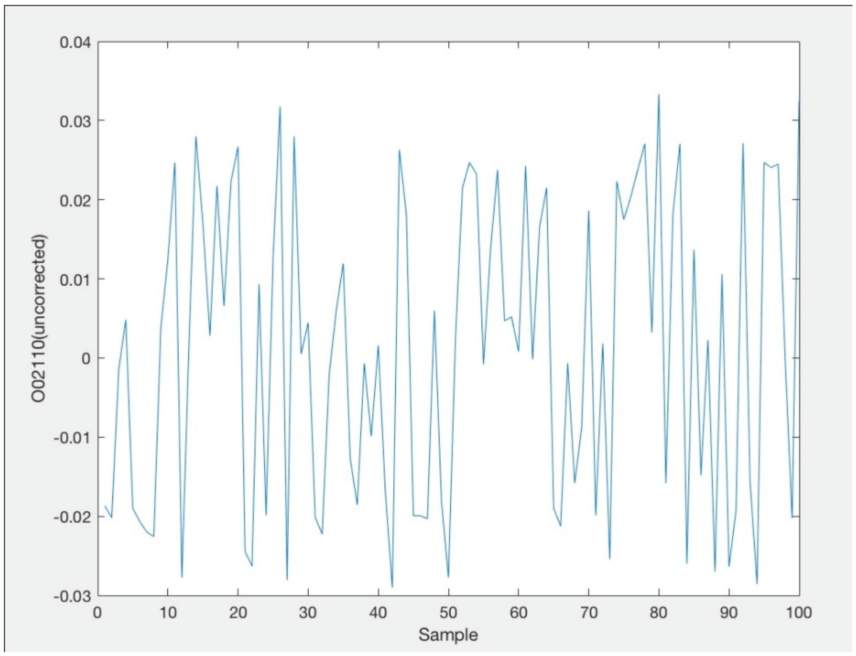

Figure 8: EP for October 2 Morning Plant 1 without heat and noise correction

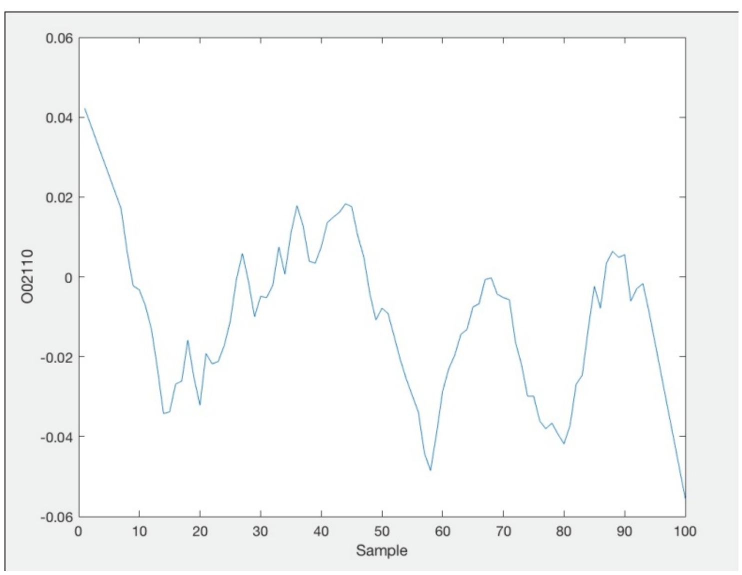

Figure 9: EP for October 2 Morning Plant 1 without heat but with noise correction

EP Readings for the same day, time period and plant have been shown below, but in case of a heat shock, without (Figure 10) and with (Figure 11) the correction algorithms applied.

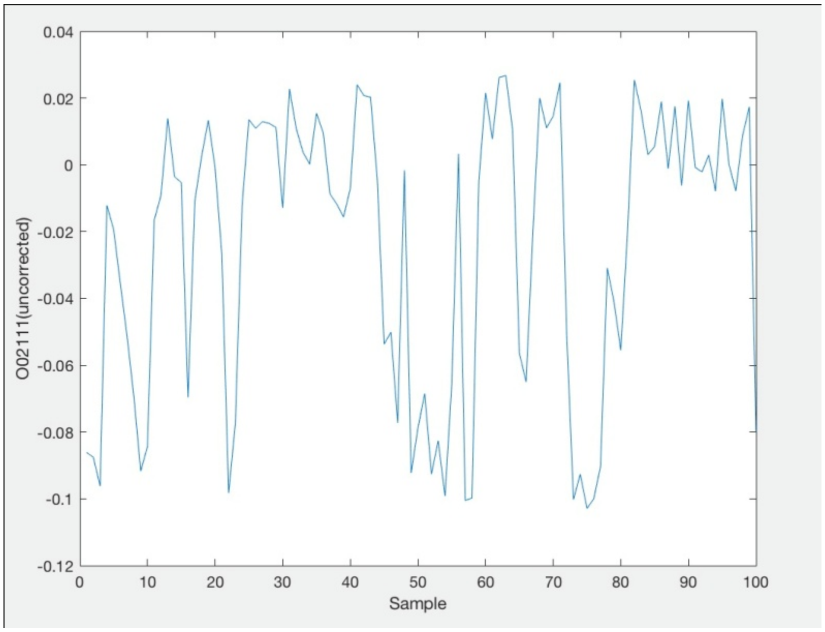

Figure 10 EP for October 2 Morning Plant 1 with heat but without noise correction

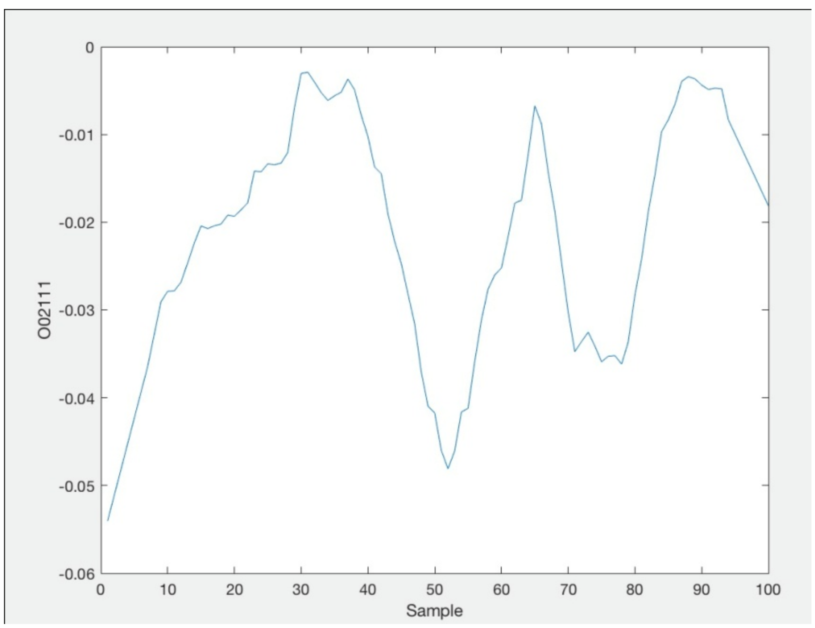

Figure 11 EP for October 2 Morning Plant 1 with heat and noise correction

A clear fall in the average data can be identified after the heat shock, with the readings before the heat shock seemingly centered slightly above 0, and the highest values of EP after the heat shock not rising above this value. Although EP generally seemed to increase in value on average as the day progressed, the data showed little equivalence when it came to values of electrical potential. However, a common pattern was that changes in the morning and afternoon time were always negative during the heat shock while changes at night always seemed to increase in the value during the project. 4 sets of the data (mean EP readings at room temperature and with the heat shock) have been shown below (Table 1) along with the difference after the heating. Evidently, difference data for the night times has been positive and higher in absolute value than for morning and afternoon. The difference data for plant 1 and plant 2, each morning, afternoon and night-time for the 10 days has been also been summarized (Table 2), and is used for the equivalence testing. A few outliers can be seen, but while morning and afternoon data seem to be decreaseing by about 0.02 to 0.04 V, night time increase in EP seems to be from 0.035 to about 0.05 V in general.

Table 1 EP Data for mornings, afternoons and nights of four selected days. The readings seemed to increase as the day progressed, but were starkly different for each plant

			Average EP value before heating/V	Average EP value after heating/V	Change in values/V
Sep-26	Plant 1	Morning	0.0036	-0.0178	-0.0214
		Afternoon	0.0316	0.0026	-0.029
		Night	-0.0073	0.0348	0.0421
	Plant 2	Morning	0.0187	-0.0116	-0.0303
		Afternoon	0.0536	0.0289	-0.0247
		Night	-0.0121	0.0274	0.0395
	.....				
Sep-30	Plant 1	Morning	-0.0021	-0.0269	-0.0248
		Afternoon	0.0645	0.0346	-0.0299
		Night	0.0633	0.0841	0.0208
	Plant 2	Morning	-0.0103	-0.0375	-0.0272
		Afternoon	0.0322	0.0014	-0.0308
		Night	0.0781	0.1184	0.0403
	.....				
Oct-02	Plant 1	Morning	0.0005759	-0.0254	-0.0198
		Afternoon	0.0524	0.0301	-0.0223
		Night	-0.0207	0.0624	0.0831
	Plant 2	Morning	-0.0312	-0.051	-0.0198
		Afternoon	0.0065	0.0013	-0.0052
		Night	0.0895	0.1237	0.0342
	.....				
Oct-04	Plant 1	Morning	0.0036	-0.021	-0.0246
		Afternoon	0.0135	-0.0079	-0.0214
		Night	0.0453	0.0865	0.0412
	Plant 2	Morning	-0.0161	-0.0416	-0.0255
		Afternoon	-0.0072	-0.0421	-0.0349
		Night	-0.0031	0.0431	0.0462

Table 2 Change in EP Value after heating on each day of the experiment for each plant

Day	Time difference	Plant 1	Plant2
Sep-25	Morning difference/V	-0.0315	-0.0213
	Afternoon Difference/V	-0.0314	-0.0421
	Night Difference/V	0.0383	0.0436
Sep-26	Morning difference/V	-0.0214	-0.0303
	Afternoon Difference/V	-0.029	-0.0247
	Night Difference/V	0.0421	0.0395
Sep-27	Morning difference/V	-0.0413	-0.0245
	Afternoon Difference/V	-0.0231	-0.0256
	Night Difference/V	0.0503	0.0402
Sep-28	Morning difference/V	-0.0205	-0.0189
	Afternoon Difference/V	-0.0371	-0.0418
	Night Difference/V	0.0437	0.0398
Sep-29	Morning difference/V	-0.0263	-0.031
	Afternoon Difference/V	-0.0201	-0.041
	Night Difference/V	0.0486	0.0472
Sep-30	Morning difference/V	-0.0248	-0.0272
	Afternoon Difference/V	-0.0299	-0.0308
	Night Difference/V	0.0208	0.0403
Oct-01	Morning difference/V	-0.0219	-0.0245
	Afternoon Difference/V	-0.0226	-0.0312
	Night Difference/V	0.0297	0.0453
Oct-02	Morning difference/V	-0.0198	-0.0198
	Afternoon Difference/V	-0.0223	-0.0052
	Night Difference/V	0.0831	0.0342
Oct-03	Morning difference/V	-0.0334	-0.0261
	Afternoon Difference/V	-0.0341	-0.0256
	Night Difference/V	0.0493	0.0452
Oct-04	Morning difference/V	-0.0246	-0.0255
	Afternoon Difference/V	-0.0214	-0.0349
	Night Difference/V	0.0412	0.0462

As indicated by the data, while mean values of the electrical signals decreased on average during the morning and the afternoon, they increased in the readings taken during the night, and increased by substantially greater amount than they decreased. After analysis with the TOST, Morning data difference seemed to be the most closely related between the two plants, as it only took 9 iterations for the loop to break- for the probability of the difference being within the increasing interval to be greater than 0.

This figure was higher, 14, for afternoon data, and 16, for night- time data. Hence, morning data changes were most constant between both the plants, followed by afternoon and night- time data.

CONCLUSION, AND FURTHER APPLICATIONS

One of the key purposes of building this device was to bring the technology close to those that may benefit from it, the farmers. The use of a simple statistical test to estimate value for a sample set, the arithmetic mean, indicates a value that should ideally be accessible to many people.

The readings taken during the experiment show promise in that EP in *Vitisvinifera* could indeed be representative of the physiological stresses it undergoes. The readings collected, on average, showed that the electrical signals at certain times of the day maychange constantly in response to equivalent stress on the plant. Indeed, in the next study, emphasis must be given on verifying these readings with more plants of the same variety. With access to industrial grade data acquisition systems, I would like to test the predictions that have been made within this research paper, and check whether the abstraction from the lower sampling rate mechanism holds. In any case, there seems potential that a grapevine plant may be ‘calibrated’ to act as a sensor of heat shocks, by collecting sufficient data, monitoring how average values change, checking to what extent these change values are constant across multiple plants of the same species, and then triggering an alarm if a sudden change of mean EP is detected in that range.

An important variable that has not measured in this study is the set of the constituent frequencies of the signal. Short Term Fourier Transform using the ‘spectrogram’ function in MATLAB was attempted, yet high frequency resolution could not be attained despite trying multiple overlap and frame length combinations.

$$STFT\{x[n]\}(m,w) = X(m,w) = \sum_{n=-\infty}^{\infty} x[n]\, w[n-m]\, e^{-jwn}$$

$$x[n] = signal;\ w[n] = windowing function$$

Low sampling rate of the ADC made it difficult to trust the values of any frequency and corresponding power/frequency readings acquired, and in a later edition, I hope to find a better data acquisition mechanism- such as a differential multiplexer mechanism with low noise addition- with a higher sampling rate to counter this issue and see possible trends of how heat shocks affect the prominent frequencies within the spectrum.

Since electrical changes have been studied in context to heat shocks, I look forward to carrying out a multivariate test in the next phase of the experiment with access to more sophisticated sensing devices for a biotic factors like humidity, soil moisture, wind velocity and so on, and experimenting with developing models through multivariate statistical tests like Principle Components Analysis and Principle Components Regression. Plant electrophysiology is a sector in its highly fundamental stages in India, and I hope that this report can help make the field more popular amongst agricultural researchers.

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References

[1] Fromm, J., and W Eschrich. "Electric signals released from roots of willow (Salix viminalis L.) change transpiration and photosynthesis ." *Plant Physiol*, 1993 : 141, 673-680.

[2] Volkov, A.G. "Green plants: electrochemical interfaces ." *Electroanalytic Chemistry*, 2000.

[3] Fromm, J., and S. Lautner. "Characteristics and functions of phloem-transmitted electrical signals in higher plants." In *Communication in Plants. Neuronal Aspects of Plant Life.* , by F. Baluska, S. Mancuso and D Volkmann. 2006.

[4] Volkov, A.G., T.C. Dunkley, S.A. Morgan, D Ruff, Y.L. Boyce, and A.J Labady. "Bio- electrochemical signaling in green plants induced by photosensory systems." *Biolelectrochemistry*, 2004: 91-94.

[5] Gil, P.M., L. Gurovich, B. Schaffer, N. García, and R Iturriaga. "Electrical signaling, stomatal conductance, ABA and ethylene content in avocado trees in response to root hypoxia." *Plant Signalling Behav.*, 2009: 4: 100-108.

[6] Stahlberg, R., and D.J. Cosgrove. "Slow wave potentials in cucumber differ in form and growth effect from those in pea seedings." *Physiol Plantarum*, 1997: 379-388.

[7] Burdon-Sanderson, J. "Note on the electrical phenomena which accompany irritation of the leaf of Dionaea muscipula." 1873: 495-496.

[8] Fromm, J., and W. Eschrich. "Transport processes in stimulated and non-stimulated leaves of Mimosa pudica. II: Energesis and transmission of seismic stimulations." 1988: 15-24.

[9] Varmuza, K., and P. Filzmoser. *Introduction to Multivariate Statistical Analysis in Chemometrics.* Boca Raton, Florida, 2009.

[10] Gil, Pilar, Torrico, Jorge Saavedra, Bruce Schaffer, Rosa Navarro, Claudia Fuentealba, and Felipe Minoletti. "Quantifying effects of irrigation and soil water content on electrical potentials in grapevines (Vitis vinifera) using multivariate statistical methods." *Scientia Horticulturae*, 2014: 71–78.

[11] Davies, E. "New functions for electrical signals in plants ." *New Phytol*, 2004: 607-610.

[12] Stahlberg, R., R.E. Cleland, and E.. Van Volkenburgh. *Slow wave potentials–a prop- agating electrical signal unique to higher plants.* Heidelberg, 2006.

[13] Xiaofei, Yan, et al. "Research progress on electrical signals in higher plants." *Science Direct, Progress in Natural Science* 19, no. 5 (2009).

[14] Hellman, EW. "Grapevine Structure and Function." *Oregon Viticulture.*

[15] JT., Levis. "ECG Diagnosis: Pulmonary Embolism." *The Permanente Journal.* 15, no. 4 (2011).

[16] LabCognition. *LabCognition.* http://www.labcognition.com/onlinehelp/en/multiplicative_scatter_correction.htm (accessed October 2, 2017).