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David Berg Utah State University

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## **Creating an Electronic Analog of a Stomatal Network**

David Berg, Faculty Mentor: David Peak

Utah State University, Logan, Department of Physics 4415 Old Main Hill, Logan, UT 84322, USA

## ABSTRACT

Stomata are microscopic variable aperture pores on the surfaces of leaves. In response to different environmental stimuli, they control the exchange rate of both water vapor and carbon dioxide between the air and the leaf interior, making their role an important determination of the status of plant life on Earth. This study is based on a recently developed stomatal model in which the aperture size is governed by vapor phase humidity. The study describes an electronic analog of the stomatal unit; in it, current models water flow, a transistor represents the variable resistance of the stomatal aperture, capacitor voltages represent turgor pressure in cellular tissue, and variable potential sources represent outside stimuli such as humidity, light, and carbon dioxide. When wired together, the combined analog represents an entire stomatal array. With this electronic circuit, experiments can be conducted on analog plants that would normally be too difficult, too timeconsuming, or too dangerous to run in a laboratory setting using biological plants.

**Keywords:** Stomata, Electronic Analog, Vaporphase Model, Operational Amplifier

## **INTRODUCTION**

The photosynthetic process is fundamentally important for the health of the biosphere and the status of the global climate. The rate at which it occurs in plant cells is dependent, in part, on the levels of water and carbon dioxide. On the epidermis of plant leaves are microscopic variable aperture pores known as stomata. These pores control the exchange of water vapor, carbon dioxide and oxygen between the air and the leaf's interior. A recent study uncovered that the stomatal aperture size is dictated by a signal from the mesophyll in response to the presence of both light and carbon dioxide [1].

In the study, experiments were conducted on the species Tradescantia pallida using conditions close those experienced in normal to environments. These conditions included using solutions between pH 4 and pH 10 and temperatures between 18°C and 30°C. Doing experiments in conditions beyond these boundaries can result in adverse effects on the plant specimens. The object of this study is to create an electronic analog of a stomatal network in order to synthesize experiments that would be performed outside these normal conditions.



Figure 1. Stomata of an isolated epidermis of T. pallida

### **METHODS**

A recent study called the Vapor-Phase Model (VPM) and performed at Utah State University by Drs. Keith Mott and David Peak found that the water potential gradients created from water transport are primarily performed in water's vapor-phase [2]. In creating an electronic analog, these water potential gradients can be modeled as an electrical current driven by voltage gradient. A potential difference is created between ground, representing the route to the root system, and  $V_w$ , representing the term  $\Delta w$  in the VPM.  $\Delta w$  is the

mol fraction of water between the leaf's interior and the air.

There are three main portions of a stoma, the pore section, the epidermal portion, and the guard cell portion. The pore opening is the primary component in creating the gradient between the inside of the leaf and the environment. Both the epidermal portion and the guard cell play important roles in the transport of water as there is an osmotic potential, modeled as additional voltage sources, in the cells. See **Figure 2** for a diagram of the analog.



**Figure 2.** Circuit making up  $\frac{1}{4}$  of a stomatal unit. Part of the circuit is shown in two other figures, the block ( $\Box$ ) portion as **Figure 3** and the triangle ( $\Delta$ ) portion as **Figure 4**.

In the analog, the different electronic components model various aspects of the plant cell structure. As previously noted, voltage sources and the potential differences they create model water potential gradients, such as V<sub>H</sub> models the currentindependent diffusive resistance of the VPM. Resistors model the resistance water has in moving between different portions, such as the resistance water would feel moving through the cell wall; R<sub>e</sub> models the resistance to liquid water flow in the xylem, Rg models the vaporto-liquid water exchange resistance between the pore and the guard cell, and rg and re model the resistance between the mesophyll and the guard cell and epidermis respectively. The capacitors Cg and Ce model turgor potential in the guard cell and epidermis respectively.

Turgor potential plays an important role in creating a water potential gradient as cells will retain water to aid in various cellular processes. In the capacitors  $C_g$  and  $C_e$ , the buildup of voltage as a function of time, *t*, is given by the equation:

$$V_c(t) = V_0(1 - e^{-t/RC})$$

where  $V_0$  is the incoming voltage, R is the resistance and C is the capacitance. Once  $V_C(t)$  approaches  $V_0$ , the current being diverted to the capacitor will instead continue to flow into the summing portion of the analog as shown by the triangle portion in **Figure 2**.

Within the guard cell branch and in the main circuit, operational amplifiers (op-amp) are used combine different branches of current. An op-amp is used for nearly ideal DC amplification. Using an op-amp in a circuit allows for signal conditioning, filtering and mathematical operations, such as addition and integration. Signals in an op-amp can be inverted (going from a positive voltage to a negative one) or non-inverted [3]. In this study, there were two primary usages of op-amps in the analog, as a summing amplifier and as a differential, or subtraction, amplifier.



**Figure 3.** The block  $(\Box)$  portion sums  $V_D$  and  $V_g$ .

An important portion of the Guard Cell branch is drawn as a block ( $\Box$ ) symbol, as shown in **Figure 3**. In this portion, an op amps are used to sum up two voltages, V<sub>D</sub> and V<sub>g</sub>, representing, respectively, water flow between the pore branch and the guard cell and light-related effects on guard cell osmotic potential. V<sub>g</sub> begins as V<sub>g0</sub>. After passing through the resistors R<sub>D</sub>, R<sub>C</sub> which models external CO<sub>2</sub> levels, and R<sub>s</sub> which models light intensity, the voltage is transformed into  $V_g$  using an op amp. After summing up  $V_D$  and  $V_g$ , a final op amp restores the voltage to the original polarity.



**Figure 4.** The triangle  $(\Delta)$  portion sums the voltages from the three branches.

The triangle section ( $\Delta$ ), as shown in **Figure 4**, symbolizes an important section of the circuit, combining the voltages of the three branches; in order from top to bottom, the epidermal branch, the guard cell branch and the pore branch. In this section, some of the resistors are multiplied by an integer *m*. The value of *m* is an integer of 2 or greater and is defined by the specific plant the experiment is modeling. For the purposes of the study, the value of 2 was assigned to *m*. The voltage out of this section then defines if the FET  $R_P$  is turned on or off, modeling the aperture being opened or closed.

It is important to note that the analog as discussed thus far makes up half of one of the two cells that create the stomatal aperture. The process for constructing a full stomatal unit will be discussed in proceeding sections.

#### **RESULTS AND EXTENSION**

In order to test the validity of the analog, it was imperative to see a result similar to one from an experiment performed on a living plant. As indicated in previous research, the opening mechanism of the aperture can be controlled by the presence and intensity of light on the guard cell [4]. Experimentally, higher intensities of light create a larger opening response than those of lesser blue light intensities.

As discussed previously, the resistor  $R_S$  in the analog represents the resistance to water movement in regards to light intensity. In this set-

up, a higher resistance in  $R_S$  will increase the output voltage. Thus, a higher  $R_S$  would then represent a greater blue light intensity.

To determine if the circuit holds true to the desired result, two resistances, 15  $\Omega$  and 30  $\Omega$  were used. Other than R<sub>s</sub>, the values for all other circuit components were held constant. Some of these values included Vw at 60 volts, V<sub>H</sub> at 10 volts, and both C<sub>e</sub> and C<sub>g</sub> at 1  $\mu$ F. Using the analog, it was determined that the time it would take to match the voltage needed to turn on the FET was 8 microsec and 5.50 microsec respectively, see **Figure 5**.



Figure 5. Voltage to turn on FET using differing values of Rs, representing light intensity. The red line is an  $R_s$  of 15 $\Omega$  and the blue line is an  $R_s$  of 30 $\Omega$ .

As seen in **Figure 5**, the resistance of  $R_s$  being doubled does not result in the FET being turned on in twice the time. As the resistance of  $R_s$ increased, the times to turn on the FET remained constant. However, as the value of  $R_s$  increased, it did result in a higher stable value. As shown in **Figure 5**, an  $R_s$  of 15 $\Omega$  resulted in a voltage that leveled out at 4 V above the on voltage while an  $R_s$  of 30 $\Omega$  resulted in a voltage that leveled out at 10 V above the on voltage. As  $R_s$  grows infinitely large, the voltage at which it levels out approaches 20 V above.

In the opposite direction, with there are values of  $R_s$  in which the circuit does not generate a voltage great enough to turn on the FET. With the values previously mentioned, it was found that an  $R_s$  of 10 $\Omega$  was just enough to turn on the FET and any resistance lower than that would fail to overcome the barrier. This data shows that the analog generates identical responses to those of biological stomata. As  $R_s$ , modeling light intensity, increased, the opening response, or the spacing between the leveling off voltage and the "On" voltage, increased. Also, it was shown that a threshold  $R_s$  value is needed to turn on the circuit.

One important note about this experiment is taking note on the time it takes to find a result. As shown in the data, the opening response to a change in  $R_s$  is in microseconds. In laboratory settings, this opening response comes on the order of minutes. An important benefit to using the analog comes from this time component, which is that the results can be found in a fraction of the time of a laboratory setting.



Figure 6. Two quarters of a stomatal unit combined, which creates one of the two cells in a stomatal unit.

The circuit described so far only makes up one half of one cell and one quarter of the entire stomatal unit. To complete the cell, two circuit components are combined together using the same  $R_e$  and  $V_w$ , as shown in **Figure 6**. By combining the two, the current is split between the two sides. Normally, the split is equal as the resistance, capacitance and voltage components on both sides of the circuit are the same. As such, the voltage needed to turn on the FET of both sides is the same and together they form a single cellular planar unit.

Combining two planar units orthogonally, as shown is **Figure 7**, creates an entire stomatal unit. Each planar portion acts a single cell. In a stomata unit, two cells work in tandem to control the aperture size. Both planar portions have the same



**Figure 7**. Two combined planar units.

potential difference as they are linked to ground and the same  $V_w$  source. In theory, one could use different resistor values for R<sub>s</sub> in each planar portion to different have a opening in piece. response each However, this result is not experimentally feasible as a fiber optic light source creates a light patch the size of an entire stomatal unit.

## **CREATING A COMPLETE ANALOG**



Figure 8. Connecting two adjacent units via a resistor.

As seen in Figure 1, the leaf surface is covered in stomatal units. As seen in experimentation, when one stoma has a change affected on it, the surrounding stomata respond in like manner. Water can be exchanged between adjacent stomatal units through the epidermal layer. Actual stomata are most certainly connected by other mechanisms besides water exchange; in this model, the simplest coupling is assumed. In order to mimic this effect in the electronic analog, each stomatal unit is linked to each other by a connecting resistor R<sub>CON</sub>, as shown in Figure 8. The value of R<sub>CON</sub> increases as the corresponding distance between two units increases.



Figure 9. A 3x3 stomatal array.

After connecting adjacent stomatal units, an entire array can be constructed. Each unit can then affect the surrounding units and create a ripple effect. Each unit has the designation (x, y). For example, in **Figure 9**, the bottom left unit would be designated as (c, a).

Creating an entire array is important in the development of the electronic analog as an actual leaf is an array of stomatal units. After preparing this array, scientists can then use it to run experiments similar to those run in the laboratory with several important benefits. First, using an electronic analog would be less time consuming than using a biological specimen. Reactions that take minutes to hours can be computed by computer within seconds. Second, experiments in adverse condition could be done with no danger to the scientist through computerized simulation.

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