NON-DESTRUCTIVE PLANT HEALTH SENSING USING ABSORPTION SPECTROSCOPY

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SUMMARY

The sensor group of the 1988 EGM 4001 class, working on NASA's Controlled Ecological Life Support Systems (CELSS) project, investigated many different plant health indicators and the technologies used to test them. The project selected by the group was to measure chlorophyll levels using absorption spectroscopy.

The spectrometer measures the amount of chlorophyll in a leaf by measuring the intensity of light of a specific wavelength that is passed through a leaf. The three wavelengths of light being used corresponded to the near-IR absorption peaks of chlorophyll a, chlorophyll b, and chlorophyll-free structures.

Interference filters, mounted on a rotating disk and placed in a beam of collimated light, are used to select a specific wavelength of light. A computer positions the disk to select the proper filter. A lens then focuses the filtered light onto the end of a fiber optic light guide, which carries the light to the detector clamp. A leaf is placed between the blades of the clamp and a photodetector is located directly opposite the end of the light guide. The computer measures the voltage produced by the detector and stores the data on a disk file for analysis.

Experimentation showed that the sensor is indeed measuring levels of chlorophyll a and b and their changes before the human eye can see any changes. The detector clamp causes little damage to the leaf and will give fairly accurate readings on similar locations on a leaf, freeing the clamp from having to remain on the same spot of a leaf for all measurements. External light affects the readings only slightly so that measurements may be taken in light or dark environments.

Future designs and experimentation will concentrate on reducing the size of the sensor and adapting it to a wider range of plants. Additional research may allow the sensor to be used in conjunction with an expert system to diagnose particular stresses and propose a treatment.

TABLE OF CONTENTS

INTRODUCTION

Problem Description

In the near future, when long term space travel becomes a reality, the need for food sources will become one of the many crucial factors controlling the duration of a space flight. The ability to grow crops in space can provide a virtually unlimited source of food and oxygen, and when astronauts count on such a closed ecological system, their survival is dependent upon the health of those crops. There is a need for some type of system that can monitor the food crops, determine if the plants are healthy or not, be able to diagnose the problem if the plants are not healthy, and finally suggest an appropriate course of action to insure the survival of the crop.

Such a system would be subdivided into two parts. The first part would encompass a way of monitoring the plants and detecting any possible problems. The second part of the system must then be able to interpret the data from the sensor and determine an appropriate course of action. An expert computer system combined with remote sensors could accomplish this task. The remote sensors would provide a way of continuously monitoring the status of the plants, and the expert system would act as a programable plant pathologist. The expert system would be able to interpret this sensory data and determine if a problem exists, and then reference its extensive knowledge and data bases to suggest a diagnosis and cure.

The success of this system is contingent upon the reliability of the remote sensor. Having a dependable automated plant health sensing system would free the astronauts from the time consuming task of visually inspecting thousands of plants on a daily basis and would not require at least one of the crew members to be an expert on plant diseases. The sensitivity of the remote sensor for detecting plant stress is important because there are many plant diseases which can harm a plant before there

are any visual signs of damage. For this reason, the development of a reliable sensing technology would be of primary importance, since one would not be able to insure the survivability of the food crops based only on infrequent visual observations.

Advances in the area of remote sensing of plant health have been slow. There are a few techniques presently being used here on earth, however, there is still a reliance upon the visual observations of agronomists and pathologists to discover and diagnose plant diseases. On earth, the losses of crops due to missed or late observations would not have the same impact as on a space mission with a limited number crops and crew members whose lives depend on those crops.

Project Description

The Sensor Project Group has selected to design a Plant Stress Sensor (PSS) which could be used to monitor the health of crops grown in space. This sensor will incorporate the use of a non-destructive form of absorption spectroscopy to measure the concentration of in vivo chlorophyll in attached plant leaves. The operation of the PSS will be completely automated through the use of a computer controller, which will also process all measurements made by the spectrometer. The development of this type of sensor is required before the entire process of automated plant health monitoring and incorporating expert systems for disease diagnosis can be implemented. The development of such a sensor would be useful for both the NASA CELSS Project as well as for the rest of the agricultural community.

Design Criteria

In designing a remote sensor to monitor the health status of plants to be grown in space, several factors must be taken into consideration. One must not only consider the wide variety of plant types and plant sicknesses, but also the special requirements for such a system to work unattended in micro gravity. Several goals were set by which to judge the merits of the various plant indicators and the technologies to sense them.

1. The indicator being sensed must give the earliest possible warning of plant stress. It must able to indicate a plant sickness (disease or deficiency) before irreversible damage has occurred.

2. The indicator must respond to a wide range of plant stresses having various types of affects upon plants. The possible sicknesses include all types of pathogen invasion, nutrient deficiencies, environmental and water stresses and physical trauma.

3. The indicator must be able to be implemented for a wide diversity of crops (or at least to the types which are being considered for space travel).

4. The sensor must not be harmful to the plant in any way that could jeopardize its growth or survival.

5. It should be an accurate and dependable indicator of plant stress. There should be no false alarms or late warnings.

6. It should give a real time indication of the health status of a plant, instead of an analysis dependent on measurements over time. In this way, the condition of many plants could be monitored more rapidly.

7. The sensor should not contaminate the growth chamber. The generation of toxic waste products or particulates would have harmful effects.

8. The system must fit within the growth chamber and be small enough to be manipulated in the growth area.

9. It must operate in microgravity and must function properly independent of orientation.

Background Information

During the Fall 1987 semester, the Sensor Project Group conducted extensive research into the area of remote sensing and plant health. The first phase of this research was to determine a good general indicator of plant health. Information was gathered from several experts in the field of plant pathology, botany, agriculture, agronomy, and food crops at the University of Florida, as well as from published research in books and professional journals. From our research, the following indicators of plant health showed to be the most promising for non-destructive sensing:

1. Changes in leaf color
2. Leaf surface temperature
3. Growth rate of the plant
4. Changes in canopy area
5. Growth rate of the flag leaf
6. Plant rigidity
7. Nutrient intake
8. Carbon dioxide intake
9. Chlorophyll levels or activity

The second phase of research was to determine the best sensing technology that could be applied towards monitoring plant health. The technologies considered where those that could be used to monitor any of the plant health indicators which are listed below:

1. Gas level/exchange monitoring
2. Infrared (IR) temperature monitoring
3. IR video imaging
4. Spectral reflectance using color IR film
5. Odor sensing
6. Ion detection/monitoring
7. Nuclear magnetic resonance
8. Electrical properties
9. Resonance frequency
10. Stimulus response monitoring
11. B & W video image processing
12. Chlorophyll level/activity determination

After much consideration and discussions with several experts, it was determined that leaf chlorophyll levels are one of the best general plant health indicators, since chlorophyll is present in all plants and that any stresses induced on the plant should affect the levels of the chlorophyll before irreversible damage occurs. Plant pathologists use the presence and pattern of chlorosis to determine whether a plant is stressed, but the human eye is sensitive to a small range of wavelengths, so it is difficult to see changes in light absortion at the particular wavelengths that would correspond to a drop in plant chlorophyll level.

After plant chlorophyll levels were chosen as the health indicator that was to be sensed, a sensing technology was now needed to measure this indicator. Extensive research showed that absorption spectroscopy would be the best method for measuring the effects of plant sicknesses on chlorophyll. A spectrometer measures the concentration of a particular element or compound by the amount of light that an unknown sample absorbs at a fixed wavelength. The Sensor Project group chose to design and construct an absorption spectrometer for measuring the concentration of chlorophyll. It was decided that it was feasible to measure the concentration of chlorophyll while it is still in the plant leaf, instead of removing a sample for destructive testing, as is traditionally done. In this way the measurement process could be greatly simplified, and the possibility of harming the plant by removing a sample would be eliminated.

Biological Aspects of Plants

Photosynthesis and Chlorophyll. During photosynthesis, water and carbon dioxide are converted into glucose and oxygen. There are two reactions that govern this transition: the light and dark reactions. In the light reaction, solar energy is collected and directed at water (a photon splits a water molecule producing ATP, NADPH, and oxygen). Then the dark reaction converts the carbon dioxide, ATP, and NADPH into glucose.

The two types of chlorophyll in the higher plants are chlorophyll a and b. The maximum absorption for these molecules ranges from 440-470 nm and 640-680 nm, respectively (Figure 1). At these wavelengths, photons are channelled into the two photosystems where the photon's energy is converted into carbohydrates. Of importance is the relative concentration of chlorophyll within the cell. The absorption value would correlate to the chlorophyll concentration, which is directly dependent on the health state of the plant. Any drastic changes in chlorophyll concentration could warn of possible harm to the plant.

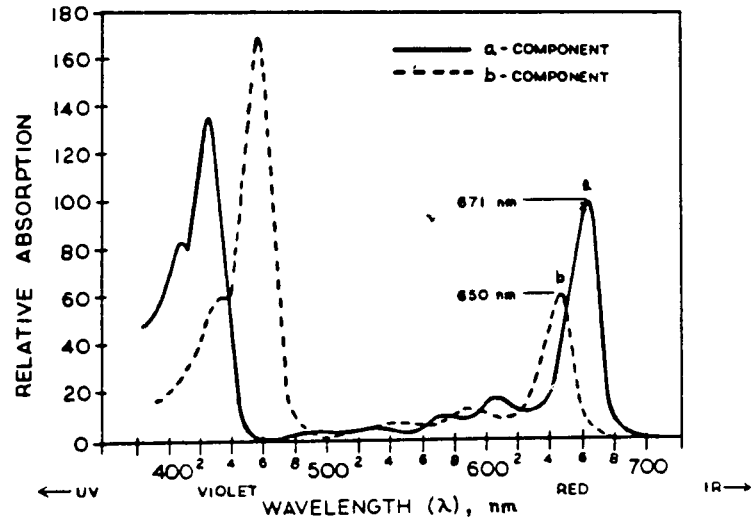


Figure 1. Absorption Spectra of Chlorophyll a & b

Chlorosis and Detectable Changes in Plant Health. Chlorosis is the loss of the green color in the plant leaf due to the decomposition of the chlorophyll in the plant cells as a result of stress. This is the cause of the yellow or dark green color in sick plants, and on initial observation of a stressed plant, chlorosis is usually the first visible warning of declining plant health [1]. Chlorosis is an early indication of a wide variety of plant stresses, the most important being improper light level, nutritional deficiencies, toxins, and pathogen invasion [1].

When present, chlorosis could appear in various locations and patterns throughout a diseased leaf. The result of improper lighting will cause an overall change in leaf color and a pattern of chlorosis related to the vein distribution with deficiencies in chlorophyll depending upon the rate of xylemic delivery [1]. A pathogen would cause localized chlorosis and these diseased spots would be detected at locations where pathogen concentrations were greatest.

Plant Stress Sensor (PSS)

The plant stress sensor uses a specially designed absorption spectrometer which non-destructively measures the amount of light being absorbed by the chlorophyll molecules at their absorption peaks. The measurement of the amount of light being absorbed by the chlorophyll relates directly to the concentration of chlorophyll in the leaf. The more chlorophyll present in a plant leaf, the more light will be absorbed by the plant at the peak wavelengths. As a plant becomes stressed, the chlorophyll molecules begin to decompose and the amount of chlorophyll begins to decrease. With this decrease in chlorophyll, the amount of light being absorbed also decreases, and this decrease can be calculated by measuring the corresponding increase in transmitted light on the other side of the leaf.

The Plant Stress Sensor (PSS) designed to accomplish this task consists of a system containing four main components (listed below), each with many subcomponents.

1. Spectrometer
2. Detector clamp
3. Interface box and power supply
4. Computer controller

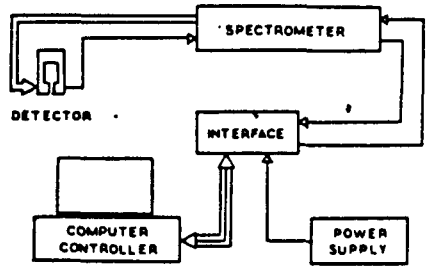


Figure 2. PSS-System Block Diagram

Spectrometer. The spectrometer module of the plant stress sensor is simple in design. It consists of a spectrometer that is configured to measure the light absorption of molecules (in this case, chlorophyll) at three predetermined wavelengths. The spectrometer delivers light of the required wavelengths to the surface of the leaf so that the intensity of the transmitted light can be measured by the photodiode on the other side (Figure 3).

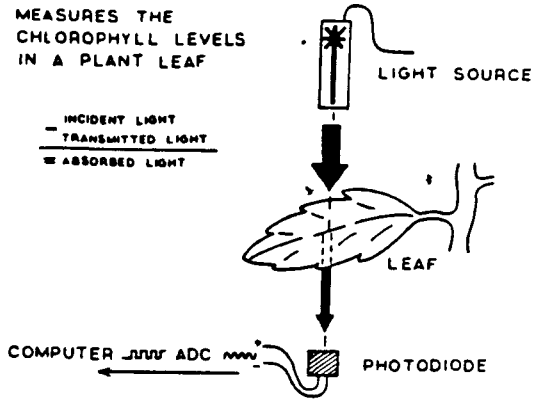


Figure 3. Absorption Spectroscopy

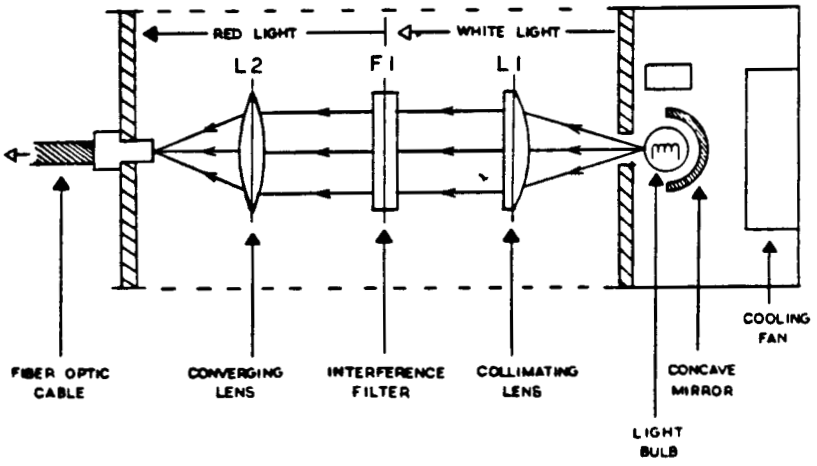


Figure 4. Spectrometer

The optical configuration of the PSS consists of five basic units: (Figure 4)

1. Small quartz-halogen bulb with rear reflector
2. Single collimating lens
3. Set of three interference filters
4. Single lens for focusing the filtered light
5. Fiber optic light guide

A 55 watt quartz-halogen bulb with rear reflector supplies white light to the interference filters. The design of the light separation section of the spectrometer is simple in comparison to existing analytic spectrometers since the absorbance of the leaf is being measured at only three wavelengths of light (650, 671, and 750 nm, each with a 10 nm half-height bandwidth). Because the measurements are being made at only these three constant wavelengths, absorption filters can be used to obtain the required wavelengths instead of the traditional prisms or interference gratings. The interference filters are mounted on a rotating disk which is driven by a stepper motor to allow the computer to position each of the filters during a measurement. The optical elements are supported on a rigid aluminum base to prevent vibrations of the optical components which would cause a variation in the measurements. A fiber optic light guide transfers the filtered light to the leaf clamp, thus allowing the actual measurement device to be relatively small. The entire

mounting system is adjustable to allow for testing of various optical configurations. A cooling fan was installed to prevent the overheating of the lamp, and all connections are removable for transportation.

Detector Clamp. The purpose of the detector clamp is to allow the filtered light to be brought to the surface of the leaf so that the light absorbance can be measured on the other side. The clamp was designed to do the following:

1. Keep the light guide and photodetector aligned, thus insuring consistent measurements.
2. Prevent the intrusion of external light.
3. Prevent damage to the leaf being measured.
4. Allow the investigator to repeatedly place the clamp over the same position on a leaf.

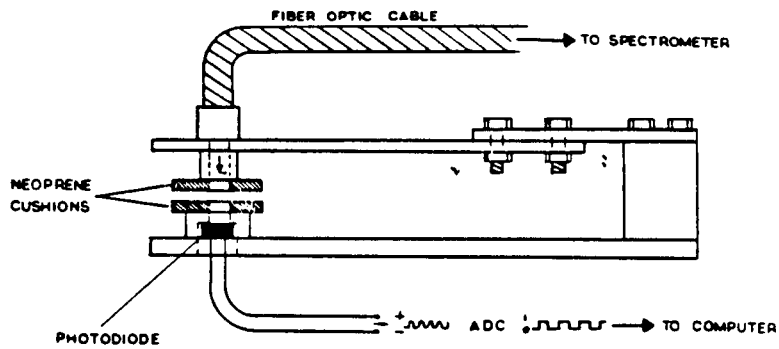


Figure 5. Detector Clamp

The detector clamp consists of a plexiglas top to hold the end of the fiber optic light guide, an aluminum bottom to house the photodetector, and a thin aluminum sheet to act as a spring between the top and bottom halves (Figure 5). The two halves of the clamp that come into contact with the leaf are covered with neoprene to prevent external light from reaching the photodetector while preventing damage to the leaf. Operation of the leaf clamp requires only one hand, thus simplifying the measurement process. A measurement is made by pulling the two clamp halves apart and inserting the portion of the leaf to be

measured over the photodiode. The clamp is then allowed to close (the spring exerts positive pressure on the two halves) and the measurement is ready to be taken.

Interface Box and Power Supply. The interface box is used as junction for all of the electrically related system components as well as an interface between the computer and the rest of the system. All data and control signals are passed through this box before being connected to the computer. Power for the interface box and electrical components within the spectrometer are also routed through the interface box.

Computer Controller. The computer controller, which is considered the 'brain' of the entire system, is used to control all of the electrical components and is also used for processing of the data. All the operations required to take a measurement of light absorption are completely automated. The computer is programmed when to take samples and sends signals to the rest of the system which causes the spectrometer to turn on, select the proper filters for measurement, input the results and process the data received from the detector and give a determination on the health of the plant.

Detailed Descriptions

The next section describes in more detail the specific operations of all the PSS subcomponents. Refer to the appendices for specific technical information or detailed schematics and drawings.

Electrical Components. The spectrometer requires six electrical components for its operation:

1. Power Supply
2. Light source
3. Stepper Motor
4. Photodetector
5. Computer Interface
6. Data Acquisition Board

Note: See the appendices for technical information, specifications, and schematic drawings.

Power Supply: The power supply used for the spectrometer is a separate unit which supplies different electrical voltages (both AC and DC) to the various components of the system. The input to the power supply is the standard 115-120 volt AC from any common house electrical outlet. This supply has 3 outputs, a 20 volt AC signal for the light, a 5 volt DC - 2.0 amp source for the stepper motor, and a 16 volt DC - 0.5 amp source for the computer interface and related electronic circuits.

Light Source: The light source consists of 4 subcomponents. A rectifying circuit, which is used to convert the 100 volt AC signal from the power supply into a 12 volt DC - 5 amp source for the light bulb. Next is the light which is a small 55 watt quartz halogen bulb that receives its power from the rectifier circuit. This bulb generates white light which can be separated into the desired color using filters. There is also a heat sensor mounted next to the light bulb which is connected to the rectifier circuit. This sensor is used to automatically disable the power to the light bulb in case it overheats. When it cools back down, it restores power to the light. The turning on or off of the light is controlled by computer software through the interface. Finally, a fan is used to cool the light bulb by drawing air from the optics portion of the spectrometer to the outside, thus removing heat from the chassis where the heat sensitive optics are located. The fan turns on when the light is

turned on and remains on for a short delay after the light is turned off to circulate air around the bulb as it cools. This time delay can be adjusted by the computer software.

Stepper Motor: The stepper motor is used to select filters for measuring the absorption of different wavelengths of light. The motor requires a 4.0 volt DC - 1.2 amp input for each step or turning of the armature. Each step is equivalent to 1.8 degrees of rotation of the motor shaft. The control of the voltage being applied to the motor is achieved through computer software and the interface.

Detector: The detector used to measure the amount of light of being absorbed by a plant leaf is a silicon PNN^{+} photodiode. This type of detector was chosen over other types of photodetectors because of its good spectral response over the wavelengths of visible light that chlorophyll a and b absorb, 650 and 671 nm respectively (Figure 6). As light strikes the surface of this photodiode, it develops a small voltage (300 - 600 mV) across its output leads. As the intensity of light striking its surface increases, so does the voltage being developed across the leads of the photodiode. This small voltage signal is sent back to the interface box where it is then channelled into the data acquisition board in the computer where this signal is amplified and processed.

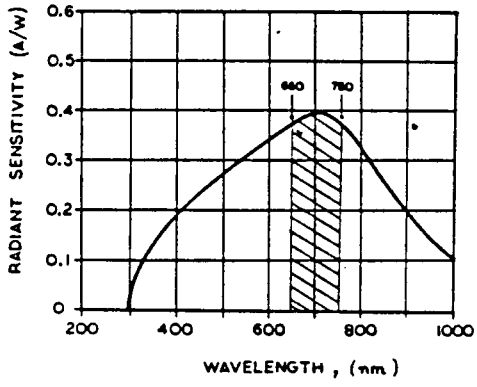


Figure 6. Spectral Response of the Photodiode

Interface Box: The Computer Interface serves as a distribution point for all of the electrical signals to and from the spectrometer, power supply, and the computer. It contains several relays which act as electrical switches used to turn the power on and off for the light, fan, and stepper motor. The actuation of the relays is achieved by sending signals from the computer which can control the time interval which a specific relay is turned on or off, thereby controlling the corresponding component which is hooked up to that relay. The control signals that are received from the computer are very weak and do not have sufficient power to turn on the relays, which require more DC current than the computer can supply. These signals must be amplified before they reach the relays, therefore the interface is equipped with a transistor circuit for each relay which is used to amplify the weaker voltage to the level required to actuate a relay. The interface box also contains LEDs (light emitting diodes) which show the status of a specific relay (on or off), as well as fuses for all the power for the system.

Data Acquisition Board: Finally, the Data Acquisition Board (DAB) which is located inside the computer serves as the Input/Output (I/O) device for all the control and data signals which are transmitted between the spectrometer and the computer through the interface box. This DAB is a commercially available expansion board (the DASCON-1 (R) produced by the MetraByte Corporation) for the IBM PC/XT or AT (R) computer. See the computer control section for more explanation of the DAB.

Computer Aspects. The operation of the spectroscope is controlled by an IBM PC/XT (R) computer running software written using GWBASIC (R). The computer is the heart of the system and allows the user to measure the health of a plant without understanding what is actually being done.

Control: When the computer software is first executed, the program initializes the DAB and loads the machine language interface required to communicate with the A/D hardware. The stepper motor is then manually incremented until it is placed in the "home" position. The "home" position is a fixed position of the filter wheel in which the white light filter is in the light path in the spectrometer. All other movements of the filter wheel are relative to this "home" position. The next step is to determine when to take a sample of light readings from the spectrometer. If a controlled experiment is being run, then the user selects a time interval for the computer to pause before automatically taking a sample and cataloging it to a disk file. If immediate health determination is desired, then a time interval of zero minutes is selected and the user presses a key on the keyboard when a health test is to be performed. For controlled experiments, the data written to the disk file is then loaded into Lotus 1-2-3 (R) for analysis.

All operations of the spectrometer are controlled by the computer. The Mechanical components directly controlled by the computer are: light source, cooling fan, and stepper motor. The light source may be manually operated by keyboard commands, or automatically operated when controlled light measurements are being taken. The fan is automatically turned on when the light is turned on, whether manually or automatically. When the light is turned off, the fan will turn off after a predetermined amount of time has elapsed. If controlled measurements are being taken, the computer also sends pulses to the stepper motor to select the proper interference filter. A total of five measurements are taken. They are: no light, 750 nm light, 650 nm light, 671 nm light, and white light. The white and no light readings are taken to store as much information as possible to help future normalizations of the data by different methods and attempt to eliminate the effects of external light interfering with the selected wavelength.

Data Acquisition: All data and control signals are connected to the computer via an analog to digital interface board (See the appendices for specifications of the DAB). The DASCON-1 board accommodates for both analog and digital input and output. Four digital outputs, two analog outputs, and one analog input are used from the DAB.

The four digital outputs control the stepper motor. The output lines drive relays which apply a large current to the motor windings because the digital lines can only support a small current load. The outputs supply either a positive voltage or a ground to each terminal of the two windings, allowing for a polarity change in the windings. Appendix A shows the sequence in which the digital outputs must be selected in order to move the motor in one direction. The sequence has both windings powered at all times to provide maximum torque. This will help reduce the chance of the disk losing alignment because of bumping or vibration during sampling.

The two analog outputs are used to control the light source and cooling fan. The analog outputs are used in the either full on or full off state, like a digital output, but were chosen over digital outputs because the full scale voltage is adjustable from 2.5 volt DC to 10.0 volt DC. These two outputs activate relays which control power to the light source and cooling fan.

The analog input is used for the photodetector. The photodiode produces a voltage between 300 mV and 600 mV which is reduced 66 percent by a 4.7/2 voltage divider. This gives a full scale input of 198 mV which is amplified by a x10 instrumentation amplifier to give a full scale input of 1.98 volt DC. This is an ideal value since the maximum input level is 2.0475 volt DC. Data is stored on the disk as bit values instead of voltage levels to help eliminate unit discrepancies.

RESULTS TO DATE

Experiments Conducted

All of the following experiments were conducted on Georgia Southern (Creole) collard plants grown in conventional planting trays. The plants reached full growth in 75 days, and the all experiments were done at about 80 days into the growth cycle. The plants were watered regularly, and the light cycle consisted of 14 hours days and 10 hour nights.

The experiments were divided into the following two areas: instrument verification and plant stress testing. Instrument verification consisted of testing the filter positioning system for alignment, alignment of the leaf clamp, and the effects of light guide positioning. After these preliminary experiments were completed and the necessary design corrections were made, the instrument was then tested on actual stressed and non-stressed plant samples.

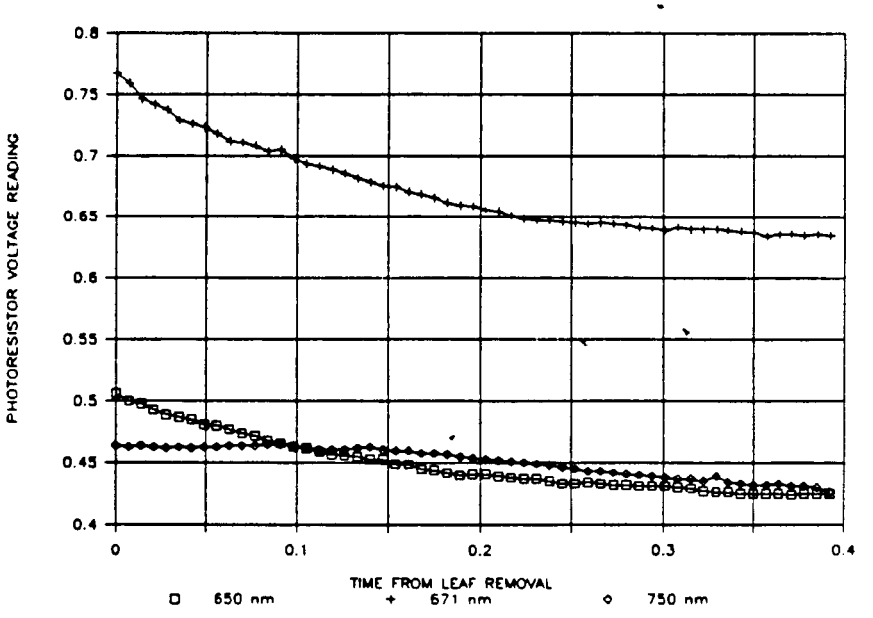


Figure 7. Spectrometric Measurements (Removed Leaf)

Cut Leaf Experiment. The purpose of the first experiment conducted was to measure transmittance light through a severed plant leaf at the three chosen wavelengths. Severing the plant leaf from the plant is a quick way of inducing stress on the leaf. It is probable that the increase in light transmittance was caused by the decreasing chlorophyll levels in the severed leaf (Figure 7). The experimental procedure was to sever the leaf from the plant and insert it into the leaf clamp for the duration of the experiment. Measurements of light transmittance were taken at regular intervals for each of the specific wavelengths over the eight hour period.

The results of this experiment show that there is a definite decrease in absorbance at the chlorophyll absorbing wavelengths. It is probable that the increase in light transmittance was caused by the decreasing chlorophyll levels in the severed leaf (Figure 7). When stress is induced by severing the leaf, the decrease in absorbtion at 671 nm is more pronounced than that at 650 nm.

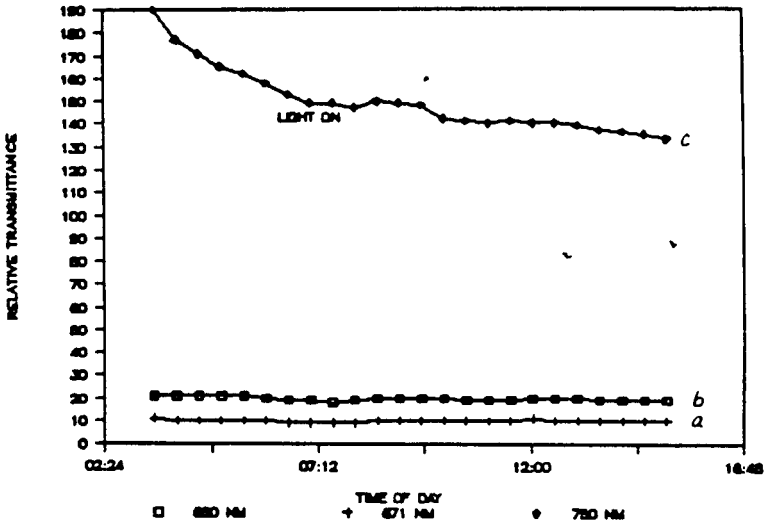


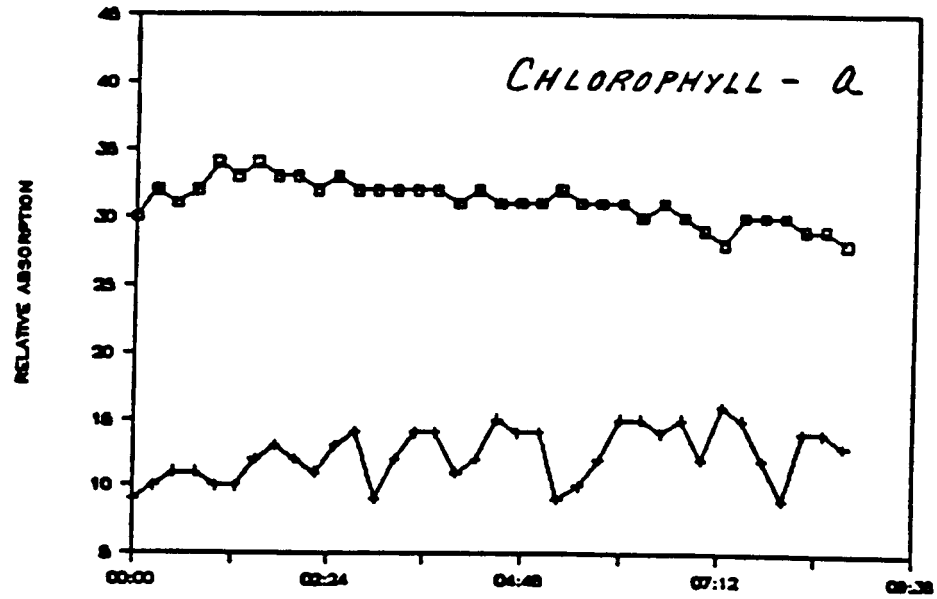
Figure 8. Light Absorption (Live Leaf)

Night to Day Transition for Live Leaf. The next experiment was conducted to determine if there was any interference caused by external light passing through the neoprene cushions. This

was done by placing the leaf clamp on a live leaf for the duration of the experiment and regularly measuring the transmittance at the three wavelengths. The experiment began at 3:00 am, and the growth chamber light came on at 7:00 am. The results of the experiment are shown in Figure 8. The graph shows that there was no measured change in the light transmittance levels for the wavelengths corresponding to the absorption peaks of chlorophyll a and b. Also, there was no visible discontinuity in the measured light intensity for any of the measured wavelengths. This seems to verify that no external light affected the measurements. Thus any changes in the measured light transmittance are a result of biological in the plant.

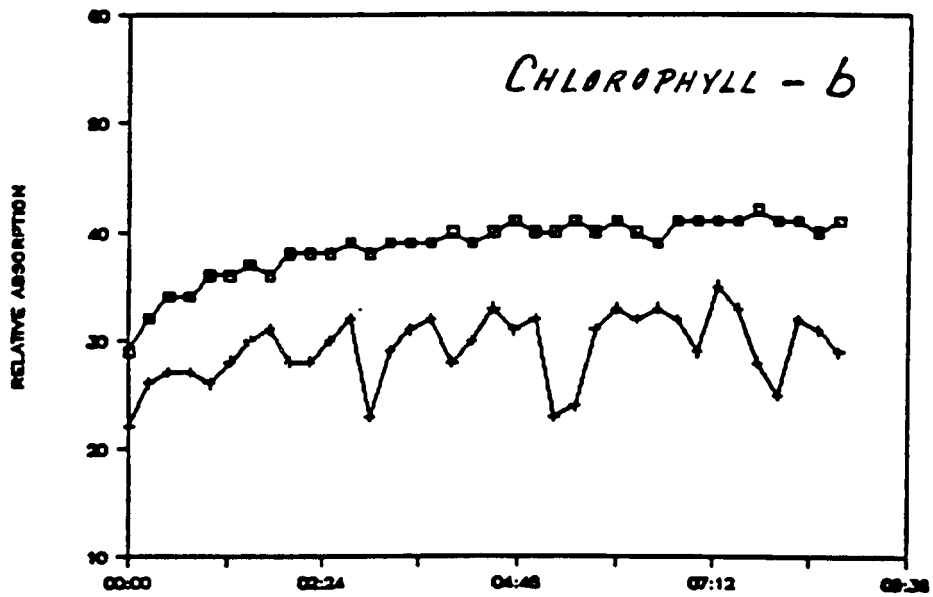
The other observation made from this experiment was that the transmittance of light at 750 nm dropped considerably over the period of the experiment. It was assumed that this drop in transmittance is due to a certain amount of stress caused by placing the clamp over the leaf for the duration of the experiment. After the experiment, inspection of the leaves revealed that the neoprene pads left small impressions on the area around the measurement spot. This cell damage could possibly cause excessive transpiration around the measurement spot, thus affecting the chlorophyll free material at this spot. This problem could possibly be alleviated by increasing the area of the neoprene pad, reducing the pressure on the leaf, or using a more resilient material for the pad.

Clamp Placement and Contact Damage. The purpose of the next two experiments was to measure the variation in transmittance induced by removing and replacing the leaf clamp in the same position. It is assumed that there will be variation in the measurement when the clamp is removed and replaced due to imperfect repositioning of the clamp. This variation could be also be caused by damage to the leaf by the clamp, by misalignment of the clamp, or by changes in the positioning of the clamp on the leaf. Since the random variation due to



Sampling Interval = 15 min

671 nm ON + 671 nm ON/OFF



Sampling Interval = 15 min

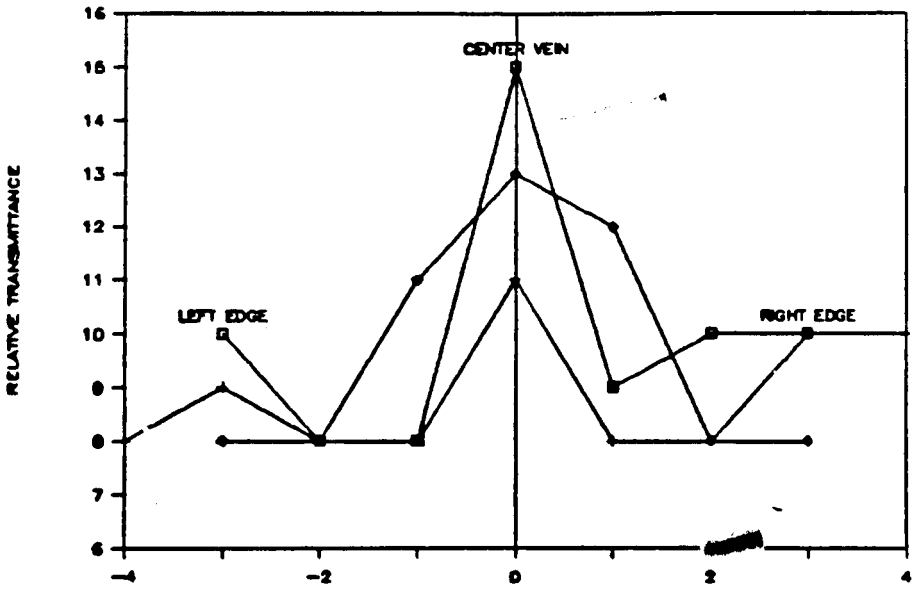
650 nm ON + 650 nm ON/OFF

Figure 9. Comparison of Sampling Techniques at 671 and 650 nm

repositioning of the clamp would have to be taken into account when deciding whether the chlorophyll level in the leaf has actually dropped, it is important that the range of variation be measured and quantified.

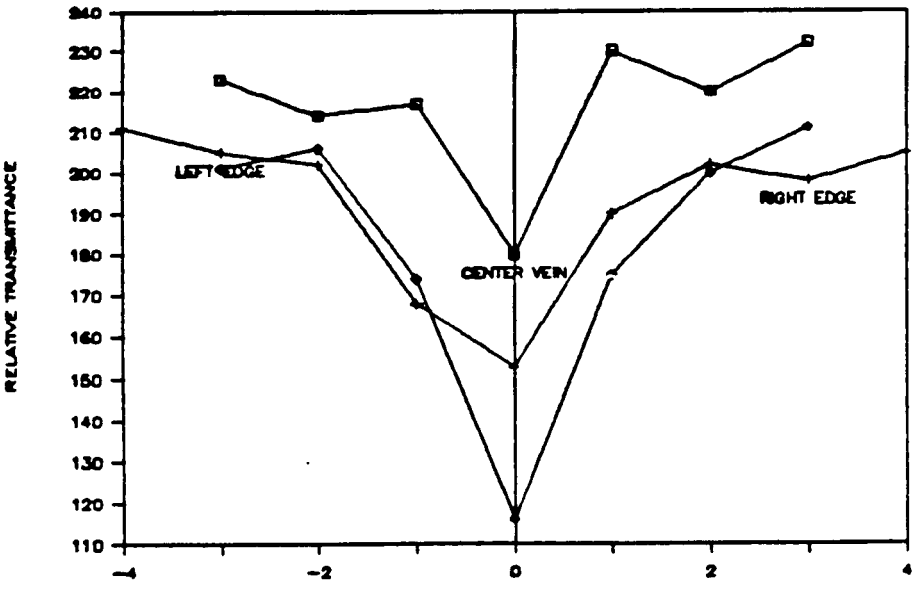
Clamp Contact Effects: The first experiment consisted of two parts; measuring the transmittance with the clamp left on and measuring transmittance with the clamp removed immediately after the measurement. In both cases the transmittance measurements were made on one third of the way from the leaf tip and stem, and one half the way between the center vein and leaf's edge. In the clamp on/off experiment, care was taken to place the clamp in the same position to minimize variations caused by actual differences in chlorophyll concentration. The measurements were made over a ten hour period. The data from the experiments are shown in Figure 9. The figure shows that there is a considerable amount of variation induced in the transmittance measurements when the clamp is taken off and repositioned. This fluctuation around the center value is most likely due to inaccurate positioning of the clamp, changes in the orientation of the leaf due to repositioning, and/or misalignment of the fiber optic cable over the photodiode due to repositioning of the clamp. Little can done to correct for fluctuations caused by the first two factors; but it may be possible to use an "area averaging" scheme, such as measuring over a larger area or "scanning" the clamp over the leaf to measure the transmittance over a larger section of the leaf.

Clamp Location: The second experiment was to test for variation in measured transmittance due to differential concentrations of chlorophyll and chlorophyll-free mater in the plant leaf. The experiment was designed to measure the transmittance at the three wavelengths as a function of the spot being measured on the leaf. A rectangular matrix was laid over the leaf by sectioning the leaf into three zones each at right



671 nm

Leaf Tip + Leaf Middle Leaf Stem



750 nm

Leaf Tip + Leaf Middle Leaf Stem

Figure 9. Light Transmittance (Live Leaf)

angles to the center vein; One section near the leaf tip, one near the center, and one close to the stem. Each of these sections was further divided into equal offsets from the center vein. Measurements were then taken at each point on the leaf. Plots of the measurements are shown in Figure 10. This figure verifies expected values. The chlorophyll concentration is fairly constant throughout the leaf itself, but the concentration is much lower it the stem of the plant, which is represented by "offset 0" for all three of the sections. On the other hand, the transmittance for the chlorophyll free structure is much lower near the vein of the leaf, and this is due to the thicker structure in this part of the leaf. These two observations coincide with visual observations; The veins of the leaf are yellow due the lower chlorophyll levels in this area, and the absorption in the veins at wavelengths that are not absorbed by chlorophyll is much higher due to the relatively thicker structure.

Physical and Chemical Stress. These experiments were run concurrently for 13 hours on three separate collards plants; One plant was a control plant, one plant had a physical stress induced upon it, and the other was induced with a chemical stress. Transmittance values were measured on three leaves for each of the plants; One young leaf at the top, one middle aged leaf, and one large leaf near the bottom of the stem were measured. Data was collected every half hour on the same spot. Several measurements were made on the stressed plants before the stress was applied to obtain a baseline value.

Control Plant: The purpose of this experiment was to measure the variation of measured transmittance due to repositioning of the leaf clamp, and to have a control to compare the stressed plant measurements with. The bottom leaf of the control plant was in an advanced stage of chlorosis due to senescence (aging). The experimental data, shown in Figure 11,

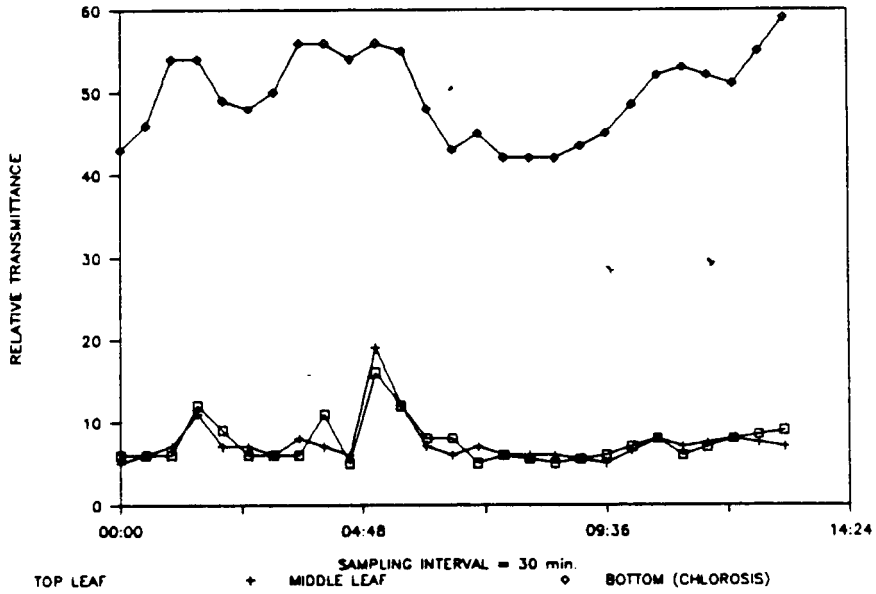


Figure 11. Control Plant: No Stress Induced (671 nm)

shows that there is no trend in the transmittance over time. One observation is that the transmittance for the bottom leaf that had begun to chlorosis was much higher that the transmittance for the two other leaves. This supports the presumption that the spectrometer is actually measuring the chlorophyll levels. Also, the transmittance for the two healthy leaves is relatively equal, and the two bottom plots of the transmittance over time are surprisingly similar.

Physical Stress: The purpose of this experiment was to find if a change in transmittance could be detected when the induced stress did not directly induce chlorosis. The physical stress was induced by rubbing the leaf surface with fine grade sand paper, thus breaking the surface cells of the leaf and allowing the water to transpire quickly from the surface. The data in Figure 12 shows no trend in changes of light absorption over the period of the experiment which suggests that variation in measurements are due to the positioning of the clamp. This is not surprising, since the chlorophyll would not be directly affected by a physical stress until the leaf begins to dehydrate.

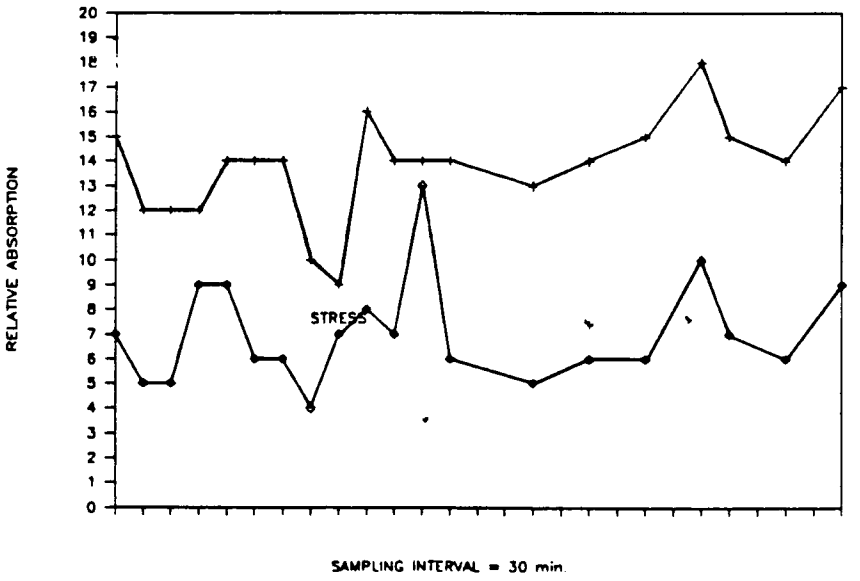


Figure 12. Physically Induced Plant Stress (650 and 671 nm)

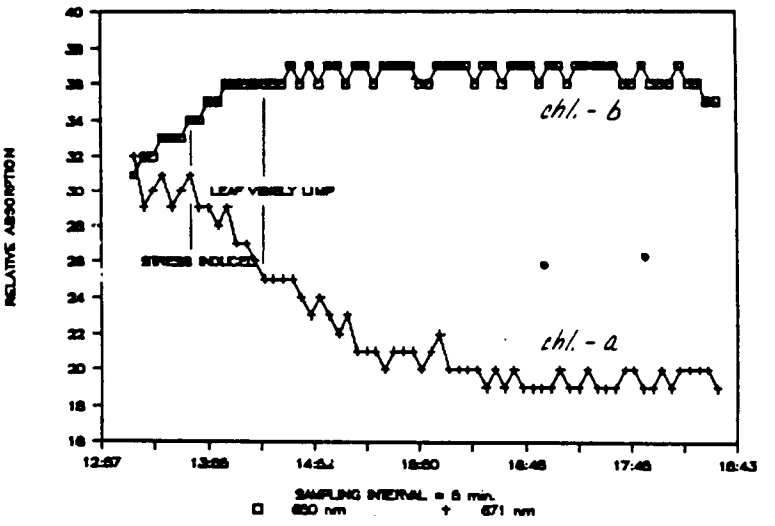


Figure 13. Chemically Induced Plant Stress (650 and 671 nm)

Chemical Stress: The purpose of this experiment was to show any changes in chlorophyll caused by introducing a toxin into the plant tissue. A leaf was severed from the plant after 2 hours of baseline measurements and the stem was placed in a solution of 30% hydrogen peroxide (H_2O_2). This chemical is a strong oxidizer which should decompose the chlorophyll and eventually bleach the leaf. The clamp was left on the leaf for the duration of the experiment to eliminate any variation due to clamp repositioning. The experimental data, shown in Figure 13, shows that there is a drastic decrease in the transmittance at 671 nm, which corresponds to the absorption peak of chlorophyll a. This is the opposite of the expected result, since the transmittance should increase drastically as the concentration of chlorophyll begins to decrease. This inconsistency from the expected result may be due to an absorption peak of H_2O_2 that corresponds to that of chlorophyll a. The transmittance at 650 nm, which corresponds to chlorophyll b, rises as expected. It was noted that the plant leaf never turned visibly yellow during the 13 hours that the experiment was run, although the leaf did become limp soon after the stress was induced.

CONCLUSION

A change in chlorophyll levels in plant leaves is one of the best indicators of common plant stresses. The Plant Stress Sensor (PSS) designed by the EGM 4001 class is a specialized spectrometer which measures in vivo chlorophyll levels in plant leaves. The measurement procedure is simple, quick, and induces little stress on the plant leaf.

Test results seem to indicate that the PSS does measure chlorophyll levels in leaves. Inducing a stress onto the plant causes an increase in the transmittance through the leaf of light of the same wavelength as the absorption peaks as chlorophyll. It is assumed that this increase in transmitted light is a result of a decrease in chlorophyll levels in the plant leaf, but further testing is necessary to verify this. A change in light transmittance through a plant leaf can be detected long before the chlorosis can be seen, and it is hoped that this change can be detected before the stress does irreversible damage to the plant.

An experiment conducted on a live unstressed plant during a "night to day" transition showed that external light levels did not affect the measurements. Thus the sensor may be used in either a day or night lighting environment without shielding the detector.

Another experiment done on stressed and unstressed plants indicates that the PSS can measure changes in light transmittance when a chlorophyll-destroying plant stress in induced onto the plant. Also, it was shown that the transmittance measured on a unstressed plant is fairly constant, except for a small amount of random variation that results from the removal and repositioning of the clamp on the plant leaf. Except for the areas around the veins and near the leaf tip, the chlorophyll level were fairly constant in the test plant type, so it does not seem important to place the clamp on the exact same spot for a series of measurements.

There are several problems associated with using the PSS to warn of changes in chlorophyll level in the plant leaf. The detector clamp also damages the leaf slightly when left on the same spot or when it is placed on the leaf carelessly. It is important that the clamp not be placed on a vein, for this will lead to an invalid measurement of leaf chlorophyll level. Another drawback with the PSS is that it may be difficult to make chlorophyll measurements for some plant types, such as wheat of lettuce, that have leaves that are incompatible with the leaf clamp design.

Also, a difficulty associated with using chlorophyll level as a stress indicator is that some stresses may not affect chlorophyll levels until the plant is incurable. Any stresses that affect only a small area of a plant may also elude result in a normal chlorophyll measurement unless the infected area happens to be measured.

RECOMMENDATIONS FOR FUTURE DEVELOPMENT

Further Experimentation

Several additional experiments will need to be conducted on the plant health sensor to determine its performance characteristics. Extensive testing will show if the sensor is performing under the specifications it was designed for.

Wavelength Selection. The sensor was designed to produce light wavelengths at 650, 671, and 750 nm to measure transmittance. A spectral scan at the delivery point of the fiber optic cable will reveal the true frequencies of light being passed to the leaf and measured by the photodetector. Much attention will be given to the 750 nm filter, as a third-order harmonic at 375 nm would be transmitting light in a region responsive to plant tissue [2]. New or additional filters may be required to filter out unwanted harmonics revealed by a spectroscopic scan.

Calibration of Spectrometer. It will be neccessary to determine the sensitivity of our spectrometer for measuring the chlorophyll transmittance values determined on a plant leaf must be compared to those achieved through known spectroscopic means. The chlorophyll would have to be isolated and the transmittance determined by a spectrophotometer for the same leaf followed by calibration of the sensor with the data obtained to insure that the spectrometer is actually measuring chlorophyll. [3]

Age Dependence. Chlorophyll distribution is related to the age of the plant leaf due to protein synthesis and degradation factors. An age versus chlorophyll concentration profile would need to be determined to correct for differences in leaf age.

Realistic Plant Stresses. Nutritional variability, toxin addition, and pathogen invasion vary the distribution of chlorosis on the plant leaf in the same manner. Each type of stress would cause a heterogeneous distribution of chlorosis and experiments would need to determine how these variables influence transmittance measurement. Some possible stresses are:

1. Water stress
2. Low light levels
3. Nutrient stress -- deficiency and toxicity
4. Physical trauma -- root, stem and leaf damage
5. Pathogen invasion

Possible Future Implementation of PSS

In an actual implementation of the PSS for the sensing of plant health in future space missions, the design of such a system would incorporate several advanced features to make it more efficient.

Robotic Clamp Manipulation. In order to minimize human interaction, the placement of the detector clamp could be performed by a robotic system. The accurate placement of the detector clamp on the plant to be measured could be done by a video pattern recognition system that could determine the position of the plant leaves. Such a system would consist of a video camera on the robotic clamping arm and an artificial intelligence system that would interpret the video signal and accurately place the clamp on the desired position on the plant to be measured.

Use of Laser as a Light Source. Traditional optical elements are prone to burn out or become misaligned, therefore a future implementation of this system would use a tunable laser system to provide the light of the required wavelengths. If future advancements allow the miniaturization of these lasers,

they could be placed on the detector clamp, thus eliminating the need for a light guide (Figure 14). Also, since a laser beam contains such a narrow bandwidth on light, external light would not have to be shielded out. This means that the detector clamp would not have to come in contact with the leaf surface.

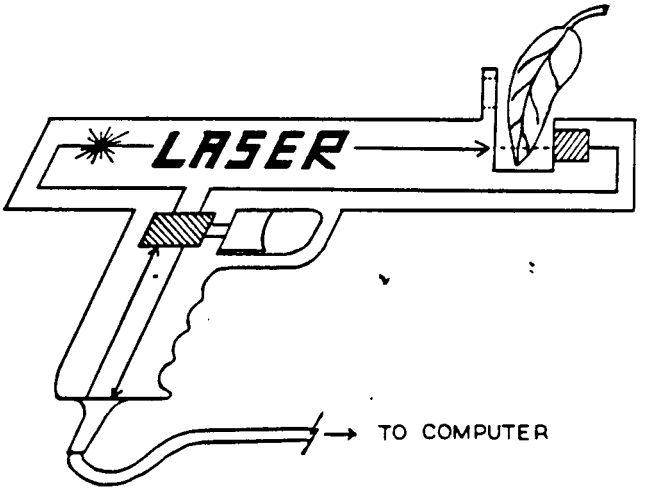


Figure 14. Future PSS

Chlorophyll level measurements would be made by "scanning" the laser across the leaf surface and measuring the transmitted light level with a photodetector that is aligned with the laser on the opposite side of the leaf.

Plant History Analysis. To increase the confidence in the decision about the health of a plant, the most recent measurement of chlorophyll levels could be compared to the past levels of that plant and to chlorophyll level of other plants of that type as the plant ages. In this way, insignificant fluctuations and/or variations due to the life cycle of the plant in the chlorophyll level can be accounted for using statistical analysis, thus decreasing the likelihood of false warnings. Using this data interpretation method will also decrease the time between the onset of a plant stress and the decision (with maximum confidence levels) that there is a problem with a plant or plants.

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7. Photodiodes: Including Si, GaAsP and GaP Photodiodes, Hamamatsu Photonics K. K., Hamamatsu City, Japan, 1987, p. 14.

8. Data Acquisition and Control, MetraByte Corp., Taunton, MA, 1983, pp. 85-89.

APPENDIX A

Optical Component List and Specifications [4,5]

Lenses.

L1: Collimating lens

Source:	Edmund Scientific	P/N: PN-A-94-257
Type:	Plano-convex	
Diameter:	30 mm	
Focal Length:	50 mm	

L2: Converging lens

Source:	Edmund Scientific	P/N: PN-A-32-243
Type:	Double convex	
Diameter:	29 mm	
Focal Length:	28 mm	

Interference Filters.

F1: Chlorophyll a

Source:	Edmund Scientific	P/N: PN-A-30-930
Wavelength:	671 nm	
Tolerance:	$\pm$ 5 nm	
Diameter:	25 mm	

F2: Chlorophyll b

Source:	Corion	P/N: S10-650-F-F241
Wavelength:	650 nm	
Tolerance:	$\pm$ 5 nm	
Diameter:	25 mm	

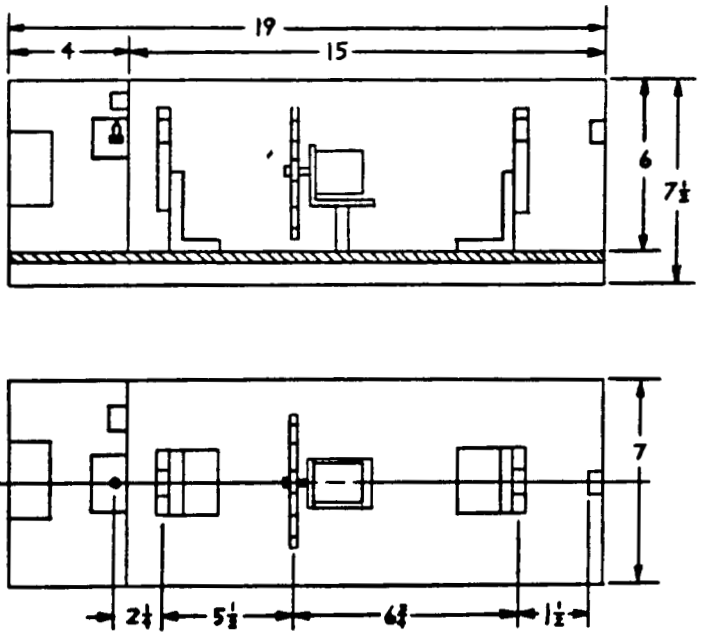
F1: Chlorophyll free structures

Source:	Corion	P/N: S10-750-F-H335
Wavelength:	750 nm	
Tolerance:	$\pm$ 5 nm	
Diameter:	25 mm	

Fiber Optic Cable.

Source:	Edmund Scientific	P/N: PN-A-40-644
Type:	Flexible bundle	
Length:	36 in	
OD:	0.125 in	

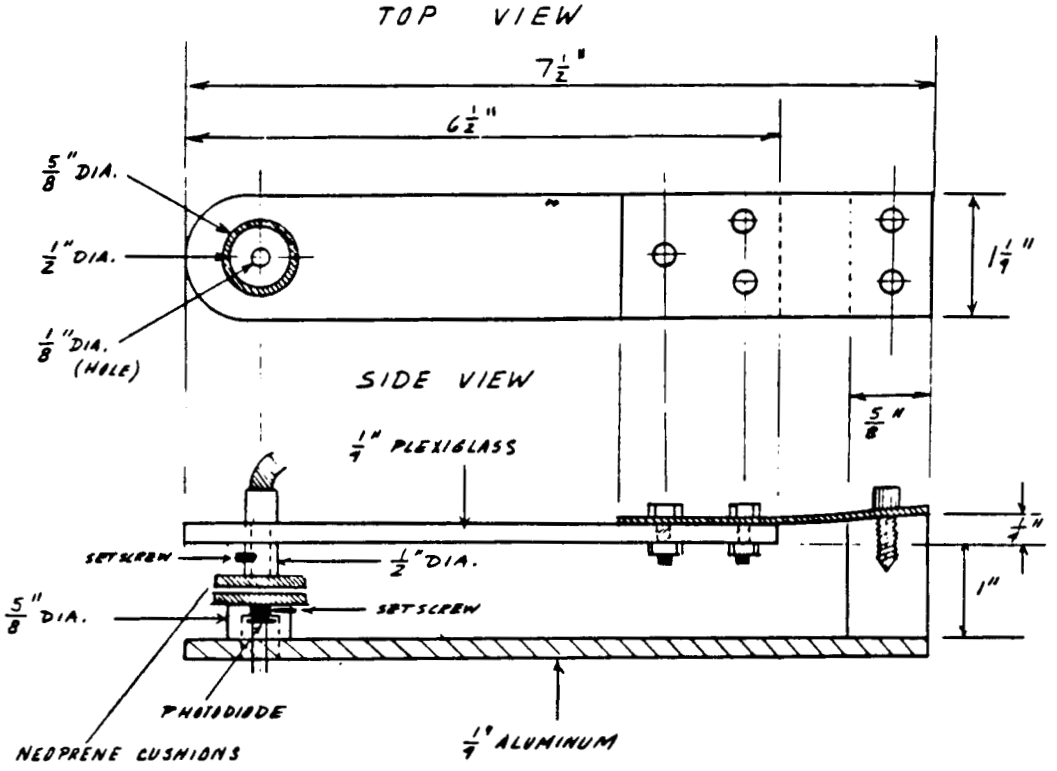
SIDE VIEW



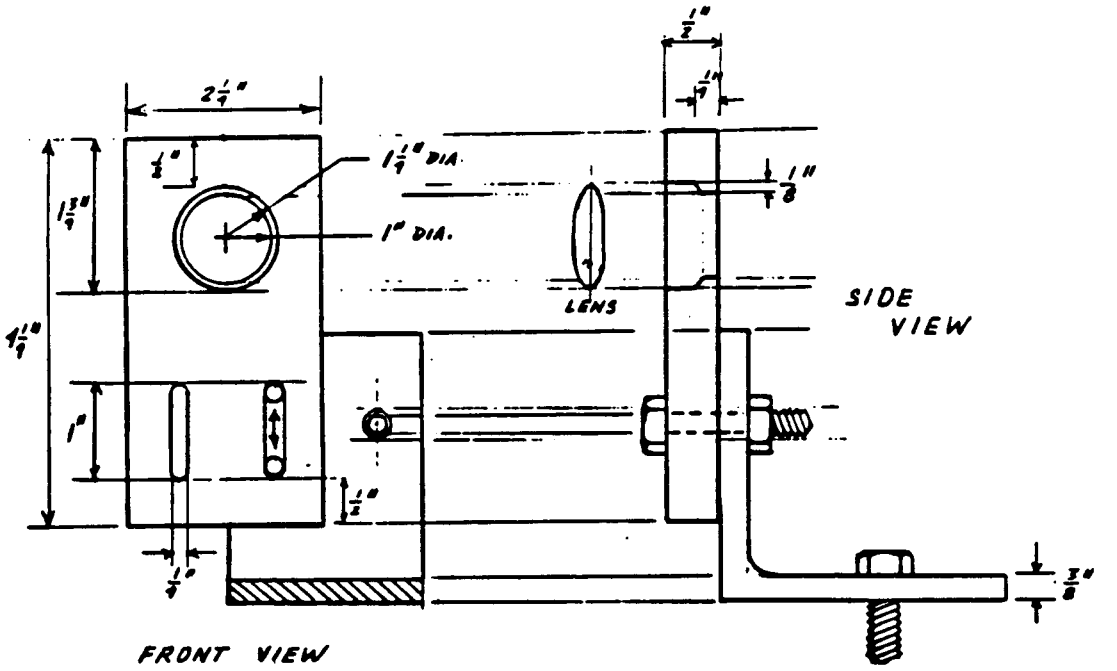
TOP VIEW

SPECTROMETER CHASSIS

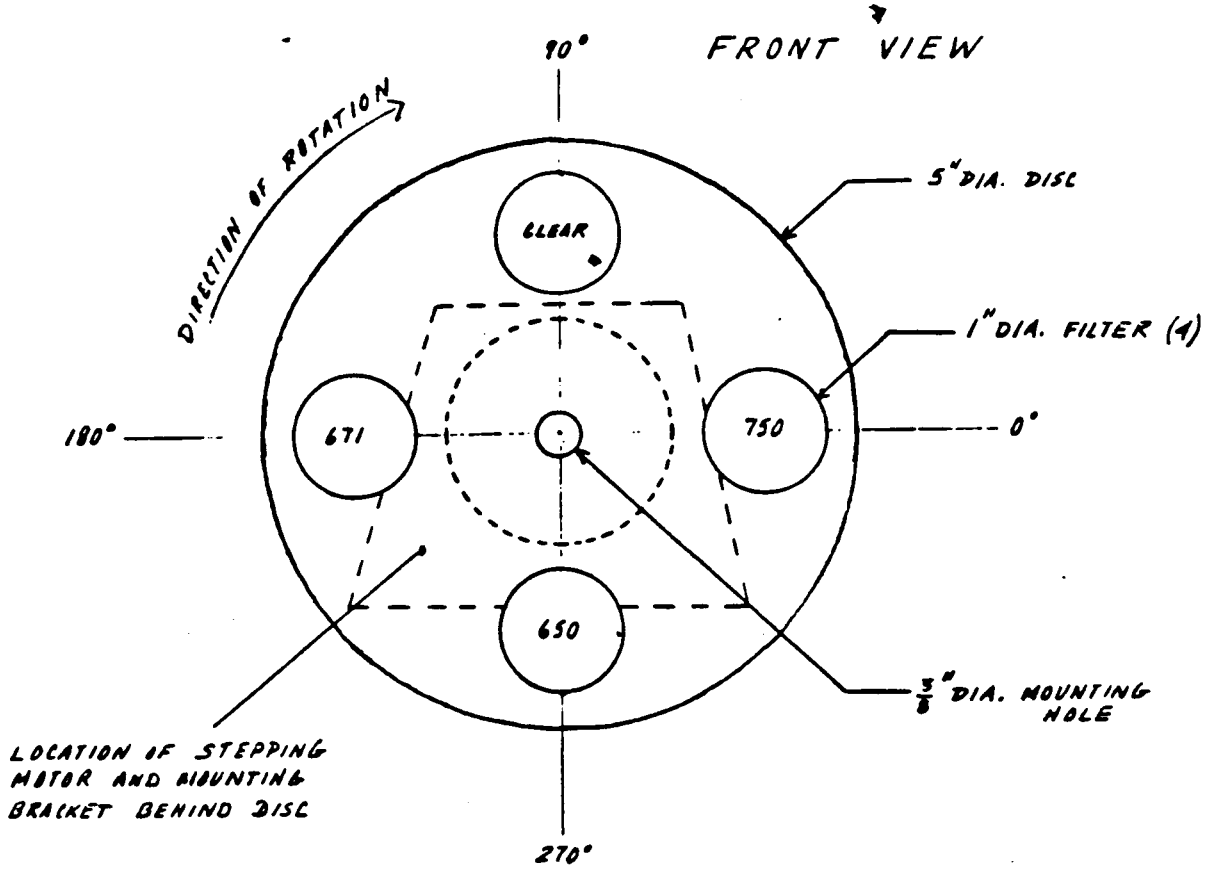
TOP VIEW



DETECTOR CLAMP



LENS HOLDER

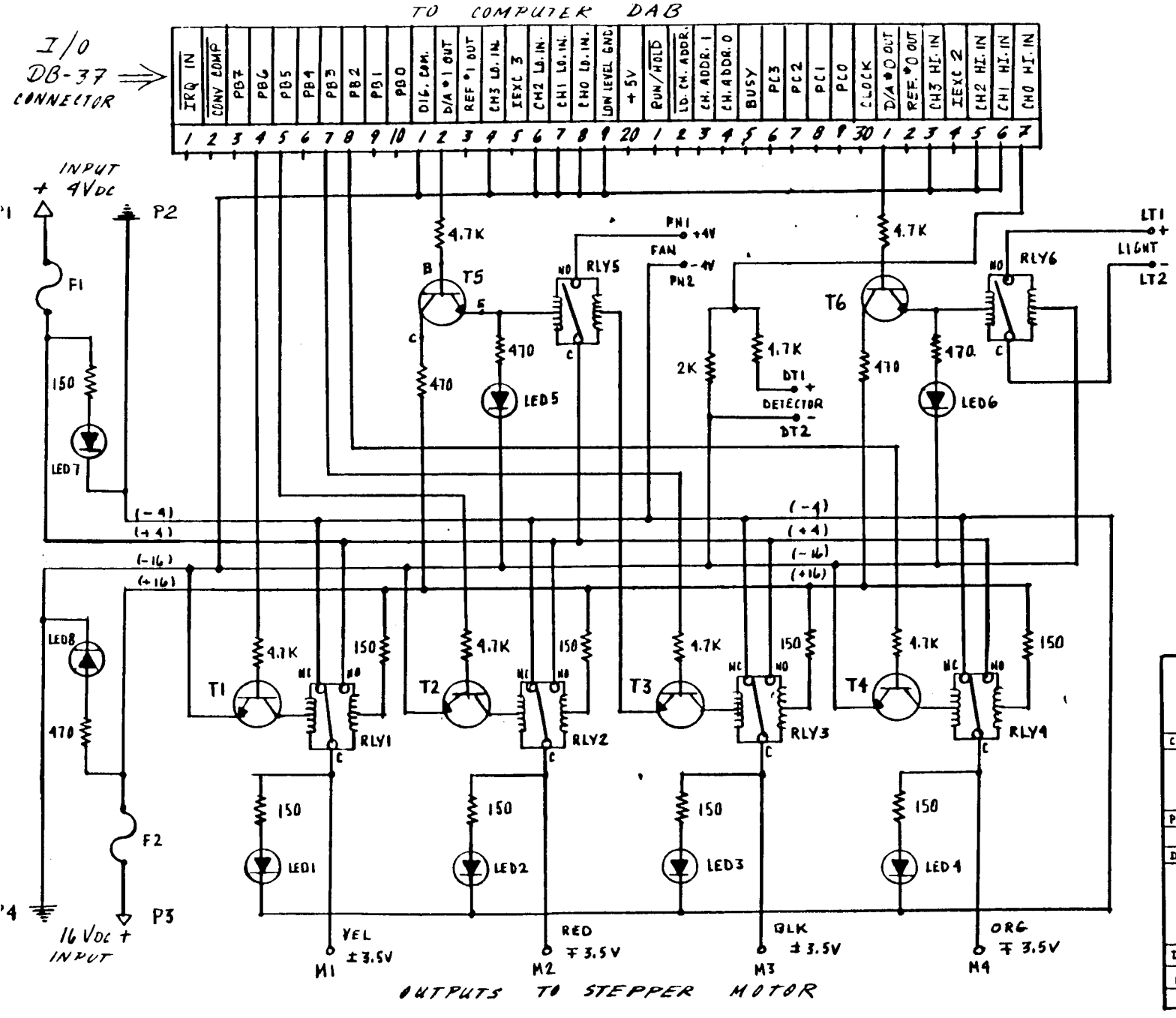


INTERFERENCE FILTER DISC

APPENDIX B

Specifications of Interface Box Electrical Components List [6]

Resistors.	(22)	1/2 watt 5% tolerance
Transistors.	(6)	T1 - T6
Type:	NPN-BJT 2N2222	
Case:	TO-18	
h_{FE}:	35 (@ V_{CE}=10 V, I_C=0.1 mA)	
I_{CMAX}:	800 mA	
V_{CEO}:	30 VDC	
V_{CBO}:	60 VDC	
V_{EBO}:	5 VDC	
Pwr. Diss.	500 mW	
Relays.	(4)	RLY1 - RLY4
Type:	SPDT subminiature	
Voltage:	5 VDC	
Resistance:	70 ohms	
Current:	72 mA	
Contact Rating:	2 A @ 125 VAC	
	(2)	RLY5 - RLY6
Type:	SPST reed	
Voltage:	5 VDC	
Resistance:	250 ohms	
Current:	20 mA	
Contact Rating:	1 A @ 125 VAC	
LEDs.	(8)	LED1 - LED8
Type:	369HHD	
Color:	red	
V_F:	1.8 VDC	
V_R:	5 VDC	
I_{FMAX}:	20 mA	
Pwr. Diss.	75 mW	
Fuses.	(2)	F1 - F2
F1:	1.5 A (@ 4 VDC)	
F2:	0.5 A (@ 16 VDC)	



UNIVERSITY OF FLORIDA
COLLEGE OF ENGINEERING
DEPARTMENT OF ENGINEERING SCIENCES

CLASS: NASA / USRA
ADVANCED SPACE MISSIONS DESIGN PROGRAM
EGM4000/1 ENGINEERING DESIGN

PROJECT: DEVELOPMENT OF A PLANT STRESS SENSOR (PSS) USING ABSORPTION SPECTROSCOPY

DRAWING TITLE:

SCHEMATIC DIAGRAM OF INTERFACE BOX

DESIGNED BY:	ARA MANUKIAN, JIM BLEDSOE
INSTRUCTOR:	DR. G. E. NEVILL
DATE:	SPRING SEMESTER, APRIL 1988

APPENDIX C

Specifications of Spectrometer Electrical Components List [7]

Photodiode.

Source:	Hamamatsu Photonics K. K.
Type:	Silicon PNN^+ photodiode
Model#:	S1226 - 5BK
Case:	TO-5
Peak Wavelength:	720 $\pm$ 50 nm
Range:	320 - 1000 nm
Effective Area:	5.7 mm^2
Size:	2.4 X 2.4 mm

Light Source.

Source:	Sylvania
P/N:	
Type:	Quartz-Halogen
Wattage:	55W
Voltage:	12 VDC

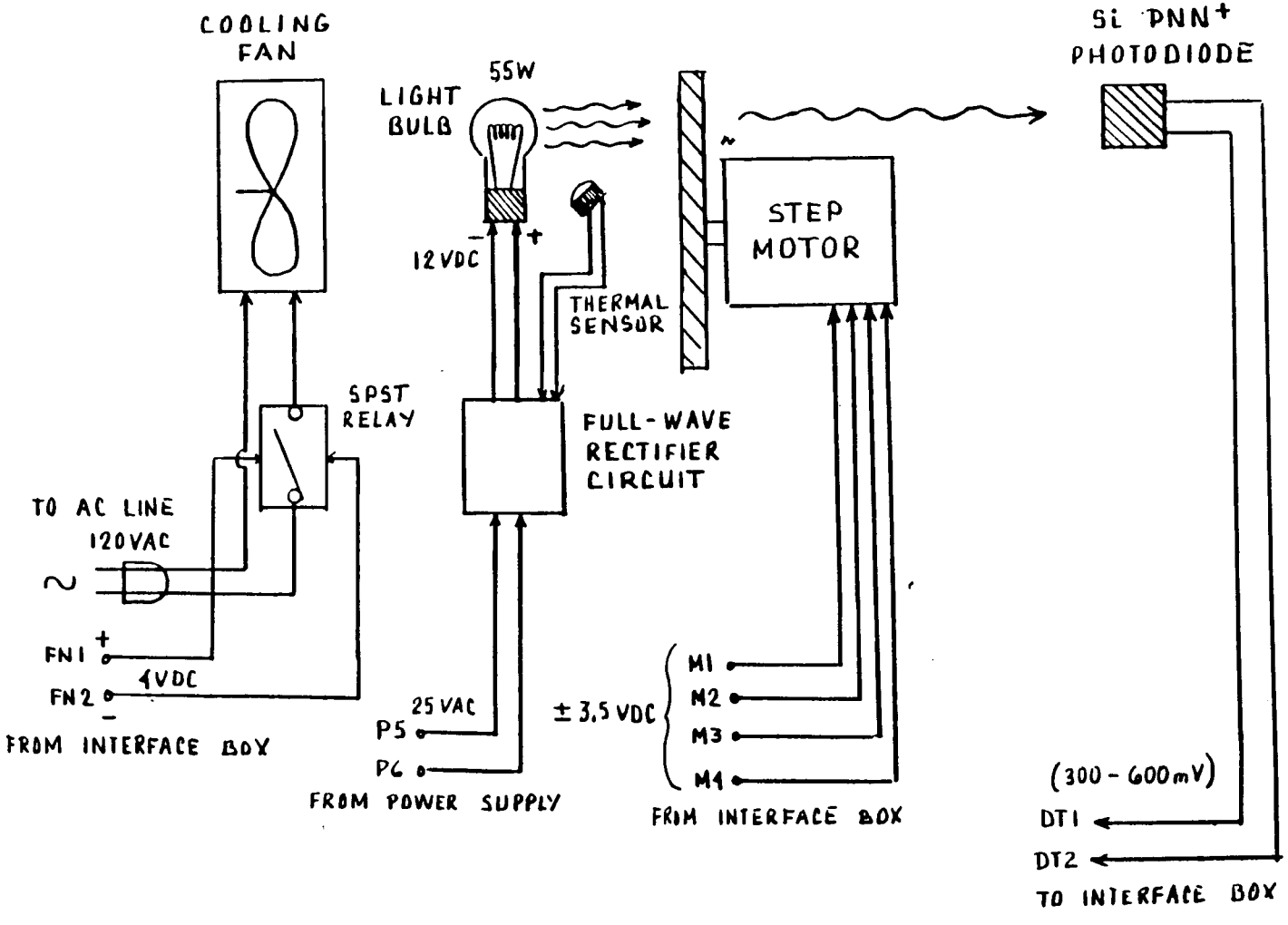
Stepper Motor.

Source:	Shinano Kenshi Co., Ltd.
Model#:	STH - 57D208
P/N:	101311 - 001 Rev. R
Steps per Rev:	200
Voltage:	3 VDC
Current:	1.2 A

Cooling Fan.

Source:	Sprite
Model#:	SU2E1
Voltage:	115 VAC
Current:	0.08 / 0.07 A
Frequency:	50 / 60 Hz
Blade Diameter:	3 in
Flow rate:	30 CFM

FUNCTIONAL SCHEMATIC FOR SPECTROMETER



UNIVERSITY OF FLORIDA COLLEGE OF ENGINEERING DEPARTMENT OF ENGINEERING SCIENCES	
CLASS:	NASA / USRA ADVANCED SPACE MISSIONS DESIGN PROGRAM EGM 4000/1 ENGINEERING DESIGN
PROJECT:	DEVELOPMENT OF A PLANT STRESS SENSOR (PSS) USING ABSORPTION SPECTROSCOPY
DRAWING TITLE	ELECTRONIC COMPONENTS OF SPECTROMETER BOX
DESIGNED BY:	ARA MANUKIAN, JIM BLEDSOE
INSTRUCTOR:	DR. G. E. NEVILL
DATE:	SPRING SEMESTER, APRIL 1988

APPENDIX D

Specifications of Data Acquisition Board [8]

Source:	MetraByte Corp.
Model:	DASCON-1

Power Consumption.

+5 V supply:	450 mA typ. / 600 mA max.
-5 V supply:	8 mA typ. / 15 mA max.
+12 V supply:	70 mA typ. / 100 mA max.
-12 V supply:	60 mA typ. / 100 mA max.

Analog Input.

Resolution:	12 bits plus sign. (0.5 mV / bit)
Accuracy:	0.01% or reading ± 1 bit
Full Scale:	± 2.0475 V
Polarity:	Automatic
Zero:	Automatic
Overvoltage:	Continuous signal channel to 120 V RMS
	5 seconds all channels to 120 V RMS
Configuration:	Full differential
Common Mode Range:	± 2 V min.
Common Mode Reject:	60 dB min., 70 dB typ.
Input Current:	1 nA max. @ 25 C
Input Filter:	Switchable on each channel
	30 dB atten. @ 60 Hz
	0.09 sec. settling time to 0.01% for FS step
Temperature:	Gain or FS, ± 25 ppm / deg. C max.
Coefficient:	Zero, ± 10 uV / deg. C max.

A/D Specification.

Type:	Integrating dual slope with auto-zero
Resolution:	12 bits plus sign
Conversion Rate:	30 conversions / sec. min.
Monotonicity:	Guaranteed over operating range
Linearity:	± 1 bit
Zero Drift:	1 uV / deg. C max.
Gain Drift:	5 ppm / deg. C max.

Instrumentation Amplifiers.

Allocated Channels:	Channel 0 and/or 1 (max. 2)
Gain Ranges:	10, 100, or 1000
Gain Error:	@ 10 -- 1.5% max. / 0.6% typ.
	@ 100 -- 0.5% max. / 0.1% typ.
	@ 1000 -- 1.5% max. / 0.4% typ.
Gain Nonlinearity:	0.01% typ. / 0.05% max.

Drift:	@ 10 -- 10 uV / deg. C typ.
	@ 100 -- 2 uV / deg. C typ.
	@ 1000 -- 1 uV / deg. C typ.
Gain Drift Coeff:	@ 10 -- 5 ppm / deg. C typ.
	@ 100 -- 5 ppm / deg. C typ.
	@ 1000 -- 15 ppm / deg. C typ.
Input Current:	10 nA max. / 2 nA typ. @ 25 C
Common Mode Range:	-2.7 V to +3.8 V min.
Common Mode Reject:	@ 10 -- 105 dB typ. / 90 dB min.
	@ 100 -- 120 dB typ. / 94 dB min.
	@ 1000 -- 130 dB typ. / 114 db min.
Overload Capacity:	120 V RMS continuous single channel

Digital I/O.

Output Lo Voltage:	0.45 V max. @ I_{sink} = 1.7 mA (1 TTL load)
Output Hi Voltage:	2.4 V min. @ I_{source} = 200 uA
Darlington Drive:	4 mA max. / 1 mA min. with R_{ext} = 750 Ohm
Input Lo Voltage:	0.8 v max. / -0.5 V min.
Input Hi Voltage:	2.0 V min. / 5.0 V max.
Input Current:	$\pm$ 10 uA max.

APPENDIX E

Single Plant Measurement Program Listing

The following appendix is a listing of the program used to take samples of a single plant without removing the clamp between samples. It is written in GWBASIC to be run on an IBM PC/XT computer with the DASCON-1 A/D board. The computer takes 8 samples for each filter and averages them before writing the three filter values plus white and no light to the disk drive for permanent storage.

```
10 SCREEN 0,0,0
20 CLEAR ,32768! : REM set up BASIC for machine language routine
30 DEF SEG = 0: SG=256*PEEK(&H511) + PEEK(&H510)
40 DASCON1 = 0: SG=(32768!/16) + SG: DEF SEG = SG
50 BLOAD "dascon1.bin",0
60 REM reset all variables
70 DASCON1 = 0: MD% = 0: CH% = 0: DIO%(0) = 0: DIO%(1) = 0
80 REM set the base address from an input file
90 OPEN "I", #1, "DASCON1.ADR": INPUT#1, BASADR%: CLOSE #1
100 REM empty paper image for simulated strip chart recorder
110 HEALTH% = 2200: DIM ARRAY%(5): DIM SAMPLE%(9)
120 REM ******************************************************
130 CLS:KEY OFF
140 REM print title and instructions for two seconds
150 LOCATE  2,16: PRINT "ENVIRONMENTAL ANALYSIS AND MONITORING
SYSTEM"
160 LOCATE  3,25: PRINT DATE$, TIME$
170 LOCATE  5,20:PRINT"Press (S) to take a sample reading"
180 LOCATE  6,17:PRINT"Press ([) and (]) to turn lamp on and off"

190 LOCATE  7,19:PRINT"Press (t) to tale five sample sequence"
200 LOCATE  8,21:PRINT"Press (m) to advance motor 4 steps"
210 LOCATE  9,24:PRINT"Press (q) to quit program"
220 LOCATE 11,1:
PRINT"=============================================================-
==================="
230 PRINT: LAMP = 0: PULSES = 0: TM = 0: MOTORPOS = 1
232 ARRAY%(1)=2^2+2^6: ARRAY%(2)=2^2+2^5: ARRAY%(3)=2^3+2^5:
ARRAY%(4)=2^3+2^6
240 LOCATE 13,1: INPUT "Enter the filename to store the data
on";FF$: IF LEN(FF$)>8 THEN GOTO 240
250 LOCATE 15,1: INPUT "Enter the experiment number for
identification";EXNUM:  IF EXNUM<0 THEN GOTO 250
260 FF$ = FF$ + ".raw"
```

```
270  OPEN "o", #1, FF$: ZERO = 0: PRINT #1, USING "JOB:
##########    ";EXNUM;:PRINT #1, LEFT$(TIME$,5),: PRINT #1,USING
"#####    #####    #####    #####
#####";ZERO;ZERO;ZERO;ZERO;ZERO: CLOSE #1
272 LOCATE 17,1: INPUT "Enter the number of minutes between
samples";MINS:  IF MINS<0 OR MINS>2000 THEN GOTO 272
273 T$=TIME$: TIM=VAL(LEFT$(T$,2))*3600 + VAL(MID$(T$,4,2))*60 +
VAL(RIGHT$(T$,2))
274 S = TIM + MINS*60: HR=INT(S/3600):
MN=INT((S-3600*INT(S/3600))/60): IF MN>59 THEN MN=MN-60: HR=HR+1
275 IF HR>23 THEN HR=HR-24
276 HR$=RIGHT$(STR$(HR),2): MN$=RIGHT$(STR$(MN),2): IF
LEFT$(HR$,1)=" " THEN HR$="0"+RIGHT$(HR$,1)
277 IF LEFT$(MN$,1)=" " THEN MN$="0"+RIGHT$(MN$,1)
278 SP$=HR$+":"+MN$+":00"
280 LOCATE 13,1: PRINT "
                                        ": LOCATE 15,1: PRINT "

      "
281 LOCATE 17,1: PRINT "
                                        "
290 REM
300 A$=INKEY$
310 IF A$="s" THEN GOTO 500
320 IF A$="q" THEN RUN 650
330 IF A$="[" THEN LAMP=4095
340 IF A$="]" THEN LAMP=0
350 IF A$="t" THEN GOSUB 980: GOTO 290
360 IF A$="m" THEN PULSES = 1
370 TM = TM - 1: IF TM<0 THEN TM = 0
380 IF LAMP>0 THEN TM = 50
390 IF PULSES>0 THEN GOSUB 730: GOTO 300
400 REM ******************************************************
410 REM ************ read the A/D channels
420 MD%=8: DIO%(0)=LAMP: DIO%(1)=0: IF TM>0 THEN DIO%(1)=4095
430 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
440 MD% = 0: CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
450 LOCATE  3,25: PRINT DATE$, TIME$
460 LOCATE 15,5: PRINT USING "Signal 1: ####.#### V     (######)
 ";DIO%(0)/2000,DIO%(0);
470 PRINT USING "    Health Deviance:#######";DIO%(0)-HEALTH%
480 LOCATE 18,4: PRINT USING "Motor position: ##      Fan
counter:########    lamp state:#######";MOTORPOS;TM;LAMP
482 IF MINS>0 THEN LOCATE 21,20: PRINT "Next sample to be taken
at ";SP$
485 IF TIME$>SP$ AND MINS>0 THEN GOSUB 980: GOTO 273
490 GOTO 300
500 REM ************ digital output section
510 REM PRINT "beginning sample.  Please do not disturb!"
520 MD% = 8 : CH% = 0
530 REM scan keyboard for any depressed keys
540 DIO%(0) = 4095: DIO%(1)=4095
```

```
550 REM call dascon board to turn lamp on
560 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
570 IF DIO%(8)>0 THEN PRINT "ERROR on digital I/O!"
580 FOR T= 1 TO 2000: NEXT T
590 REM see if they want to quit
600 GOSUB 860: REM take 8 samples
610 DIO%(0) = 0: MD% = 8: DIO%(1)=4095: TM=50
620 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
630 IF DIO%(8)>0 THEN PRINT "ERROR on digital I/O!"
640 GOTO 290
650 REM type run 1000 if program crashes with the lamp still on
660 LOCATE 24,1: PRINT "turning lamp off!"
670 DIM DIO%(8)
680 MD% = 7 : CH% = 0
690 DIO%(0) = 0
700  OPEN "I", #1, "DASCON1.ADR": INPUT#1, BASADR%: CLOSE #1
710 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
720 END
730
REM*************************************************************-
********
740 REM**   Turn motor PULSES*4 pulses.  If LSTATE>0 then turn
fan on.    **
750
REM*************************************************************-
********
760 LSTATE=0: IF TM>0 THEN LSTATE=1
800 FOR JJ%=1 TO PULSES: MD%=9: CH%=0: DIO%(0)=LSTATE +
ARRAY%(MOTORPOS)
810 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
820 REM FOR T=1 TO 30: NEXT T
825 MOTORPOS = MOTORPOS + 1: IF MOTORPOS>4 THEN MOTORPOS =
MOTORPOS - 4
827 LOCATE 18,4: PRINT USING "Motor position: ##";MOTORPOS
830 NEXT JJ%
840 PULSES=0
850 RETURN
860
REM*************************************************************-
********
870 REM**   Take 8 samples from channel 1 and average them.
DIO%(0)     **
880
REM*************************************************************-
********
890 SUM%= 0: FOR N = 1 TO 8
900 MD% = 0: CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
910 FOR T = 1 TO 250: NEXT T: REM pause for 1/4 sec
920 SUM% = SUM%+ DIO%(0): NEXT N
930 REM LOCATE 24,1: PRINT "  8 samples averaged for channel 1
=";SUM%/2000/8
940 REM PRINT TIME$;:PRINT USING"   voltage:##.###  (#####)
```

```
Health Deviance:#####";SUM%/2000/8,SUM%/8,(SUM%/8)-HEALTH%
950 REM LPRINT TIME$;:LPRINT USING"   voltage:##.###  (#####)
Health Deviance:#####";SUM%/2000/8,SUM%/8,(SUM%/8)-HEALTH%
960 REM IF ABS(SUM%/8-HEALTH%)<50 THEN PRINT "This plant is
healthy!!!!!!!!!" ELSE PRINT "ATTENTION!    This plant is
dying!!!!!"
970 DIO%(0) = SUM%/8: RETURN
980
REM*************************************************************-
********
990 REM**   Take all five samples and put in array samples()
       **
1000
REM*************************************************************-
********
1010 LOCATE 25,17: PRINT "Beginning sample.  Please do not
disturb!"
1020 MD% = 8 : CH% = 0
1030 DIO%(0) = 4095: DIO%(1) = 4095
1040 REM call dascon board to turn lamp on
1050 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
1060 IF DIO%(8)>0 THEN PRINT "ERROR on digital I/O!"
1070 MD% = 9 : CH% = 0: DIO%(0) = 1
1080 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
1090 IF DIO%(8)>0 THEN PRINT "ERROR on digital I/O!"
1100 TM = 10
1110 FOR T= 1 TO 2000: NEXT T
1120 PULSES=24:GOSUB 730:FOR T=1 TO 1   :NEXT T:GOSUB
860:SAMPLE%(1)=DIO%(0)
1130 PULSES=24:GOSUB 730:FOR T=1 TO 1   :NEXT T:GOSUB
860:SAMPLE%(2)=DIO%(0)
1140 PULSES=54:GOSUB 730:FOR T=1 TO 1   :NEXT T:GOSUB
860:SAMPLE%(3)=DIO%(0)
1150 PULSES=49:GOSUB 730:FOR T=1 TO 1   :NEXT T:GOSUB
860:SAMPLE%(4)=DIO%(0)
1160 PULSES=49:GOSUB 730:FOR T=1 TO 1   :NEXT T:GOSUB
860:SAMPLE%(5)=DIO%(0)
1170 LOCATE 23,1: PRINT TIME$,:PRINT USING "BLK:#####
750:#####    650:#####    671:#####
WHT:#####";SAMPLE%(1),SAMPLE%(2),SAMPLE%(3),SAMPLE%(4),SAMPLE%(5)

1180 REM  DIO%(0) = 0: MD% = 7
1190 REM  CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
1200 REM  IF DIO%(8)>0 THEN PRINT "ERROR on digital I/O!"
1210 MD%=9: CH%=0: DIO%(0)=0
1220 CALL DASCON1(MD%, CH%, DIO%(ZERO%), DIO%(ONE%), BASADR%)
1230 TM=50: GOSUB 1250
1235 LOCATE 25,1: PRINT "
                                        "
1240 RETURN
1250
REM*************************************************************-
```

```
********
1260 REM**   Append data to disk file for experiment#, time, 5
values.      **
1270
REM*************************************************************-
********
1280 OPEN "I", #1, FF$: OPEN "O", #2, "tmp.dat"
1290 INPUT #1, L$: PRINT #2, L$: IF EOF(1)=0 THEN GOTO 1290
1300 CLOSE #1
1310 PRINT #2, USING "JOB: ##########    ";EXNUM;:PRINT #2,
LEFT$(TIME$,5),: PRINT #2,USING "#####    #####    #####    #####
  #####";SAMPLE%(1);SAMPLE%(2);SAMPLE%(3);SAMPLE%(4);SAMPLE%(5)
1320 CLOSE #2: KILL FF$: NAME "tmp.dat" AS FF$
1330 RETURN
```

APPENDIX F

Multiple Plant Measurement Program Listing

The following appendix is a listing of the program used to take samples of up to ten different plants by removing the clamp after each sample. It is written in GWBASIC to be run on an IBM PC/XT computer with the DASCON-1 A/D board. The computer takes 8 samples for each filter and averages them before writing the three filter values plus white and no light to a disk file for permanent storage. The sampling intervals for each plant are independent of each other and each plant has a separate data file for permanent storage.

```
10 SCREEN 0,0,0
20 CLEAR ,32768! : REM set up BASIC for machine language routine
30 DEF SEG = 0: SG=256*PEEK(&H511) + PEEK(&H510)
40 DASCON1 = 0: SG=(32768!/16) + SG: DEF SEG = SG
50 BLOAD "dascon1.bin",0
60 REM reset all variables
70 DASCON1 = 0: MD% = 0: CH% = 0: DIO%(0) = 0: DIO%(1) = 0
80 REM set the base address from an input file
90 OPEN "I", #1, "DASCON1.ADR": INPUT#1, BASADR%: CLOSE #1
100 REM empty paper image for simulated strip chart recorder
110 HEALTH% = 2200: DIM ARRAY%(5): DIM SAMPLE%(9)
120 REM ************************************************
130 CLS:KEY OFF
140 REM print title and instructions for two seconds
150 LOCATE  2,16: PRINT "ENVIRONMENTAL ANALYSIS AND MONITORING
SYSTEM"
160 LOCATE  3,25: PRINT DATE$, TIME$
180 LOCATE  5,17:PRINT"Press ([) and (]) to turn lamp on and off"

190 LOCATE  6,19:PRINT"Press (t) to tale five sample sequence"
200 LOCATE  7,21:PRINT"Press (m) to advance motor 4 steps"
205 LOCATE  8,20:PRINT"Press (1) - (0) to sample plant 1-10"
210 LOCATE  9,24:PRINT"Press (q) to quit program"
220 LOCATE 11,1:
PRINT"=============================================================-
==================="
230 PRINT: LAMP = 0: PULSES = 0: TM = 0: MOTORPOS = 1: DIM
FF$(10), EXNUM(10)
240 ARRAY%(1)=2^2+2^6: ARRAY%(2)=2^2+2^5: ARRAY%(3)=2^3+2^5:
ARRAY%(4)=2^3+2^6
245 LOCATE 13,1: INPUT "Enter the number of independent
```

```
experiments"; NUMEXPS: IF NUMEXPS>10 OR NUMEXPS<0 THEN GOTO 245
247 IF NUMEXPS=0 THEN MINS=0: GOTO 300
248 FOR EXNUM = 1 TO NUMEXPS
250 LOCATE 15,1: PRINT USING "Enter the filename to store
experiment ## data on";EXNUM;: INPUT FF$(EXNUM): IF
LEN(FF$(EXNUM))>8 THEN GOTO 250
260 LOCATE 17,1: PRINT USING "Enter the experiment number for
identification of experiment ##";EXNUM;: INPUT EXIDNUM(EXNUM):
IF EXIDNUM(EXNUM)<0 THEN GOTO 260
270 FF$(EXNUM) = FF$(EXNUM) + ".raw"
280  OPEN "o", #1, FF$(EXNUM): ZERO = 0: PRINT #1, USING "JOB:
##########    ";EXIDNUM(EXNUM);:PRINT #1, LEFT$(TIME$,5),: PRINT
#1,USING "#####    #####    #####    #####
#####";ZERO;ZERO;ZERO;ZERO;ZERO: CLOSE #1
285 NEXT EXNUM
290 LOCATE 19,1: INPUT "Enter the number of minutes between
samples";MINS:  IF MINS<0 OR MINS>2000 THEN GOTO 290
300 T$=TIME$: TIM=VAL(LEFT$(T$,2))*3600 + VAL(MID$(T$,4,2))*60 +
VAL(RIGHT$(T$,2))
310 S = TIM + MINS*60: HR=INT(S/3600):
MN=INT((S-3600*INT(S/3600))/60): IF MN>59 THEN MN=MN-60: HR=HR+1
320 IF HR>23 THEN HR=HR-24
330 HR$=RIGHT$(STR$(HR),2): MN$=RIGHT$(STR$(MN),2): IF
LEFT$(HR$,1)=" " THEN HR$="0"+RIGHT$(HR$,1)
340 IF LEFT$(MN$,1)=" " THEN MN$="0"+RIGHT$(MN$,1)
350 SP$=HR$+":"+MN$+":00"
360 LOCATE 13,1: PRINT "
                                        ": LOCATE 15,1: PRINT "

      "
370 LOCATE 17,1: PRINT "
                                        ": LOCATE 19,1: PRINT "

      "
380 REM
************************************************************-
******
382 REM **  begin the keyboard scanning and wait for sample time
to arive   **
385 REM
************************************************************-
******
390 A$=INKEY$
410 IF A$="q" THEN RUN 760
420 IF A$="[" THEN LAMP=4095
430 IF A$="]" THEN LAMP=0
440 IF A$="1" THEN EXNUM = 1: GOSUB 1080: GOTO 380
441 IF A$="2" THEN EXNUM = 2: GOSUB 1080: GOTO 380
442 IF A$="3" THEN EXNUM = 3: GOSUB 1080: GOTO 380
443 IF A$="4" THEN EXNUM = 4: GOSUB 1080: GOTO 380
444 IF A$="5" THEN EXNUM = 5: GOSUB 1080: GOTO 380
445 IF A$="6" THEN EXNUM = 6: GOSUB 1080: GOTO 380
```

```
446 IF A$="7" THEN EXNUM = 7: GOSUB 1080: GOTO 380
447 IF A$="8" THEN EXNUM = 8: GOSUB 1080: GOTO 380
448 IF A$="9" THEN EXNUM = 9: GOSUB 1080: GOTO 380
449 IF A$="0" THEN EXNUM = 10: GOSUB 1080: GOTO 380
450 IF A$="t" THEN EXNUM = 99: GOSUB 1080: GOTO 380
455 IF A$="m" THEN PULSES = 1
460 TM = TM - 1: IF TM<0 THEN TM = 0
470 IF LAMP>0 THEN TM = 50
480 IF PULSES>0 THEN GOSUB 840: GOTO 390
490 REM ************************************************************
500 REM ************ read the A/D channels
510 MD%=8: DIO%(0)=LAMP: DIO%(1)=0: IF TM>0 THEN DIO%(1)=4095
520 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
530 MD% = 0: CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
540 LOCATE  3,25: PRINT DATE$, TIME$
550 LOCATE 15,5: PRINT USING "Signal 1: ####.#### V     (######)
 ";DIO%(0)/2000,DIO%(0);
560 PRINT USING "    Health Deviance:#######";DIO%(0)-HEALTH%
570 LOCATE 18,4: PRINT USING "Motor position: ##      Fan
counter:########    lamp state:#######";MOTORPOS;TM;LAMP
580 IF MINS>0 THEN LOCATE 21,20: PRINT "Next sample to be taken
at ";SP$
590 IF TIME$>SP$ AND MINS>0 THEN GOSUB 5000: GOTO 300
600 GOTO 390
750 REM
************************************************************-
******
760 REM type run 1000 if program crashes with the lamp still on
765 REM
************************************************************-
******
770 LOCATE 24,1: PRINT "turning lamp off!"
780 DIM DIO%(8)
790 MD% = 7 : CH% = 0
800 DIO%(0) = 0
810  OPEN "I", #1, "DASCON1.ADR": INPUT#1, BASADR%: CLOSE #1
820 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
830 END
840
REM************************************************************-
********
850 REM**   Turn motor PULSES*4 pulses.  If LSTATE>0 then turn
fan on.    **
860
REM************************************************************-
********
870 LSTATE=0: IF TM>0 THEN LSTATE=1
880 FOR JJ%=1 TO PULSES: MD%=9: CH%=0: DIO%(0)=LSTATE +
ARRAY%(MOTORPOS)
890 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
900 REM FOR T=1 TO 30: NEXT T
910 MOTORPOS = MOTORPOS + 1: IF MOTORPOS>4 THEN MOTORPOS =
```

```
MOTORPOS - 4
920 LOCATE 18,4: PRINT USING "Motor position: ##";MOTORPOS
930 NEXT JJ%
940 PULSES=0
950 RETURN
960
REM*************************************************************-
********
970 REM**   Take 8 samples from channel 1 and average them.
DIO%(0)     **
980
REM*************************************************************-
********
990 SUM%= 0: FOR N = 1 TO 8
1000 MD% = 0: CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
1010 FOR T = 1 TO 250: NEXT T: REM pause for 1/4 sec
1020 SUM% = SUM%+ DIO%(0): NEXT N
1030 REM LOCATE 24,1: PRINT "  8 samples averaged for channel 1
=";SUM%/2000/8
1040 REM PRINT TIME$;:PRINT USING"   voltage:##.###  (#####)
Health Deviance:#####";SUM%/2000/8,SUM%/8,(SUM%/8)-HEALTH%
1050 REM LPRINT TIME$;:LPRINT USING"   voltage:##.###  (#####)
 Health Deviance:#####";SUM%/2000/8,SUM%/8,(SUM%/8)-HEALTH%
1070 DIO%(0) = SUM%/8: RETURN
1080
REM*************************************************************-
********
1090 REM**   Take all five samples and put in array samples()
        **
1100
REM*************************************************************-
********
1110 LOCATE 23,17: PRINT "Beginning sample.  Please do not
disturb!"
1115 IF EXNUM<11 THEN LOCATE 25,18: PRINT USING "Taking samples
for experiment ##";EXNUM
1120 MD% = 8 : CH% = 0
1130 DIO%(0) = 4095: DIO%(1) = 4095
1140 REM call dascon board to turn lamp on
1150 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
1160 IF DIO%(8)>0 THEN PRINT "ERROR on digital I/O!"
1170 MD% = 9 : CH% = 0: DIO%(0) = 1
1180 CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
1190 IF DIO%(8)>0 THEN PRINT "ERROR on digital I/O!"
1200 TM = 10
1210 FOR T= 1 TO 2000: NEXT T
1220 PULSES=24:GOSUB 840:FOR T=1 TO 1   :NEXT T:GOSUB
960:SAMPLE%(1)=DIO%(0)
1230 PULSES=24:GOSUB 840:FOR T=1 TO 1   :NEXT T:GOSUB
960:SAMPLE%(2)=DIO%(0)
1240 PULSES=54:GOSUB 840:FOR T=1 TO 1   :NEXT T:GOSUB
960:SAMPLE%(3)=DIO%(0)
```

```
1250 PULSES=49:GOSUB 840:FOR T=1 TO 1    :NEXT T:GOSUB
960:SAMPLE%(4)=DIO%(0)
1260 PULSES=49:GOSUB 840:FOR T=1 TO 1    :NEXT T:GOSUB
960:SAMPLE%(5)=DIO%(0)
1270 LOCATE 23,1: PRINT TIME$,:PRINT USING "BLK:#####
750:#####    650:#####    671:#####
WHT:#####";SAMPLE%(1),SAMPLE%(2),SAMPLE%(3),SAMPLE%(4),SAMPLE%(5)

1280 REM  DIO%(0) = 0: MD% = 7
1290 REM  CALL DASCON1(MD%, CH%, DIO%(0), DIO%(1), BASADR%)
1300 REM  IF DIO%(8)>0 THEN PRINT "ERROR on digital I/O!"
1310 MD%=9: CH%=0: DIO%(0)=0
1320 CALL DASCON1(MD%, CH%, DIO%(ZERO%), DIO%(ONE%), BASADR%)
1330 TM=50: IF EXNUM<=NUMEXPS THEN GOSUB 1360
1340 LOCATE 25,1: PRINT "
                                                "

1350 RETURN
1360
REM*************************************************************-
********
1370 REM**   Append data to disk file for experiment#, time, 5
values.      **
1380
REM*************************************************************-
********
1385 LOCATE 25,5: PRINT "Saving the data to file ";FF$(EXNUM);:
PRINT USING " for experiment ##";EXNUM
1390 OPEN "I", #1, FF$(EXNUM): OPEN "O", #2, "tmp.dat"
1400 INPUT #1, L$: PRINT #2, L$: IF EOF(1)=0 THEN GOTO 1400
1410 CLOSE #1
1420 PRINT #2, USING "JOB: ##########    ";EXIDNUM(EXNUM);:PRINT
#2, LEFT$(TIME$,5),: PRINT #2,USING "#####    #####    #####
#####
#####";SAMPLE%(1);SAMPLE%(2);SAMPLE%(3);SAMPLE%(4);SAMPLE%(5)
1430 CLOSE #2: KILL FF$(EXNUM): NAME "tmp.dat" AS FF$(EXNUM)
1440 RETURN
5000
REM*************************************************************-
********
5010 REM**   Sound an alarm until the user presses a key
             **
5020
REM*************************************************************-
********
5025 LOCATE 25,1: PRINT "Yo dude!  wakeup!  time to take all the
samples!!!"
5027 FREQ = 100
5029 FREQ = FREQ*1.2
5030 SOUND FREQ,.4
5040 FOR T=1 TO 20: NEXT T
5050 A$=INKEY$: IF A$<>"" THEN GOTO 5055
5052 IF FREQ<2000 THEN GOTO 5029
```

```
5053 GOTO 5027
5055 LOCATE 25,1: PRINT "
                                                "
5060 RETURN
```