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FINAL TECHNICAL REPORT

Potato Respirometer Experiment
S061

(NASA Contract NAS 9-9302)

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TABLE OF CONTENTS

TABLE OF CONTENTS
(Concluded)

LIST OF FIGURES

LIST OF TABLES

1.0 PROGRAM REVIEW

This program has included two separate and distinct phases. A brief review of program history is included herein to provide perspective on the body of this final report.

Several years ago efforts were begun under NASA Headquarters contract NASw-870 to develop prototype hardware for measurement of the rhythmic dian behavior exhibited by the oxidative metabolism of potato sprouts. The basis for this feasibility study was two-fold: it was based on the discovery and subsequent exhaustive evaluation of this phenomena by Dr. Frank A. Brown of Northwestern University and it was selected as an experiment potentially suitable for study on a manned space flight. The objective of the original study was to determine if a small, light weight, space flight compatible respirometer could be designed. Reference (1) describes the results of this effort.

Subsequent to the development of an operating respirometer, COMAR responded to an announcement by NASA of space flight opportunities for biological experiments on the SKYLAB series. The proposal[2] was accepted and work begun on the development of flight hardware in April, 1969. Hardware development proceeded through the Critical Design Review Stage and into flight hardware production.

In September 1971, NASA Headquarters convened a new biological experiments evaluation committee to review the rationale and hardware status of the flight status biological experiments.

Basically, some members of the committee questioned the validity of the conclusions derived from Dr. Brown's work and also questioned the capability of the flight hardware to measure the phenomena, if it existed. A special 30 day test program was subsequently conducted at the University of California, Davis, to evaluate the hardware and to verify, if possible, the presence of a rhythm. No conclusive biorhythmic data was obtained.(3) As a result, the Potato Respiration Experiment was removed from flight status in early 1971 and redirected. The revised objective of the program was to develop a laboratory facility suitable for measurement of the oxidative metabolism of potato sprouts using the flight hardware units and to then determine if the dian rhythmicity did indeed exist. The first objective has been met; i.e., a complete laboratory has been designed and built. Due to lack of sufficient time and funds, data has not yet been acquired. However, it is now anticipated that the system will be made available to a University for the purpose of continuing this important study.

Since the advent of the earth satellite, the biological sciences community has been interested in examining the behavior of biological specimens in the space environment. The influence of several phenomena on the biological specimen that are unique to space flight can be studied including zero gravity and removal from all other geophysical environments including the 24 hour rhythmic environment. The physiologic and psychologic behavior of man when exposed to the unique space flight environment is of great interest. Less complex biological specimens can be used to study aspects of basic physiologic functions. Also, there are important, basic biological phenomena thought to be related to the interaction of the geosphere with biological specimens. Thus, in these areas, space flight provides a unique opportunity to better understand the biosphere. The S061 developmental and experimental program plan satisfied the latter two objectives.

2.0 BIOLOGICAL REVIEW

Biorhythmicity in behavioral and physiologic activity has long been recognized. As techniques in disciplined observation were improved, it soon became clear that all living things demonstrated some degree of periodicity and that if enough data were collected about nearly any biologic parameter and proper analytic methods were used, a periodicity appeared to be present. The periods displayed by organisms in nature reflect the principal cyclic events of their immediate environment. Thus, such periods as solar day, tidal, synodic month, and annual have been observed in a wide variety of plants and animals.

When organisms are carefully shielded from the rhythmic variations in all ordinary factors of their environment, such as light, temperature, humidity, and tides, their behavior still remains rhythmic. Under such controlled conditions, however, a rhythmic duplicity commonly becomes evident. Precise, mean, environmentally related frequencies (dian, lunar, monthly, annual) remain present, but in addition, the overt daily or tidal physiological patterns may now display regular periods differing somewhat from the moon- or sun-related ones. In other words, the events in the recurring patterns occur regularly either a little earlier or a little later each day. These odd-lengthened periods have been the reason for coining the term circadian (circa = about; dian = day, or, "about a day long") to emphasize the fact that these rhythms actually differ in period from the dian twenty-four hour rhythm. In either circumstance, some kind of "biological clock" appears to be operating, "timing" these regularly recurrent behaviors.

How these "biologic clocks" are "driven" or "set" is not now completely understood and constitutes one of the central problems in biology today. Two alternative hypotheses have been offered to explain these rhythmic functions.

The first is that each organism is timed by an autonomous, endogenous, intrinsic, inherited and intracellular oscillator. This hypothesis postulates that each organism has within it its own "clock" by which it "sets" its rhythmic activities, and that this evolutionarily related internal clock matches closely and adaptively the major geophysical periods.

The second hypothesis postulates that each organism's timing is dependent upon exogenous, extrinsic inputs from the pervasive, subtle, rhythmic, geophysical forces from which the organisms are never fully shielded. Such forces include the Earth's gravity, and its magnetic, electrostatic and electromagnetic fields.

Until recently it has been impossible to place an organism of known biorhythm completely outside the Earth's rhythmic sphere of influence. Now with the opportunity to use space as a laboratory such removal became possible. The S061 experiment was based upon the need to deprive an organism of significant information about its rhythmic geophysical environment, and measure the effect of such deprivation upon the observed biologic rhythm. The potato Solanum Tuberosum (whose dian oxidative metabolic rhythm seemed well-defined) was selected for a flight test to separate an organism from the Earth's geophysical influences. If the rhythm remained unaltered, the intrinsic hypothesis would appear more tenable; if the rhythm was altered, the extrinsic clock hypothesis would be supported. In short, any behavioral change would be informative, and provide insight into this crucial biologic question whose answer has so many important basic and applied connotations.

A more complete biological review and a list of appropriate references can be found in references (2) and (4).

3.0 RESPIROMETER DESCRIPTION

The respirometer developed under this contract is designed to measure the oxygen consumption rate of a respiring potato sprout. In addition, the respirometer is configured to hold constant the extraneous environmental variables of light, temperature, pressure, relative humidity and gaseous partial pressure to the greatest extent possible. The variation in temperature and pressure is limited to a range which does not influence, insofar as is known, rhythmic biologic behavior.

Severe mechanical and operational limitations on respirometer design were imposed by the unique laboratory environment of SKYLAB. Accordingly, particular emphasis was placed on safety and reliability. In addition, the permissable experiment envelope was constrained by the particular mounting area selected in the Command Module. Respirometer depth was limited by the crash envelope of the adjacent crew couch. Complete mounting and location drawings are available which delineate the size and shape limitations. The general respirometer design specifications are given in Table I.

The respirometer consists of two major subassemblies (see Figure 1): a life support system called the Potato Respirator Unit (PRU) and the electronics system referred to as the Data Storage Unit (DSU).

The PRU includes the life support system and the data acquisition transducers. The enclosure acts as a closed thermal environment. The doors permit crew access for specimen inspection and recovery and permit activation of the back-up specimen if required.

The location of the major components of the PRU are shown in Figure 2 and include: two potato chambers, a high pressure oxygen storage bottle, a solenoid valve and controlled flow orifice, a manifold block, the potato chamber selector valve, the absolute pressure transducer, two temperature probes and a heater unit.

Two potato chambers, hermetically independent, provide the controlled environment for the 7.5 gram sprouting tubers. The nominal earth ambient atmosphere is obtained simply by closing the chamber in the laboratory. As shown in Figure 3, the potato

TABLE I

General Data - Potato Respiration Experiment

1.0	Life Support System Parameters		
	a.	Pressure:	758.0 ± 0.1 to 762.0 ± 0.1 mm Hg
	b.	Temperature:	23.3 ± 0.5°C
	c.	Humidity:	≥ 90% R.H.
	d.	Gaseous Environment:	Nominal Earth Ambient Atmosphere
	e.	Illumination:	Complete Darkness
2.0	Experiment Duration:		56 Days
3.0	Structure		
	a.	Weight	5100 grams excluding spacecraft mounting brackets and electrical connector
	b.	Envelope:	
		1. Mounting Bracket:	58.0 x 27.9 x 1.88 cm
		2. PRU:	17.8 x 15.8 x 9.85 cm
		3. DSU:	15.2 x 21.6 x 10.2 cm

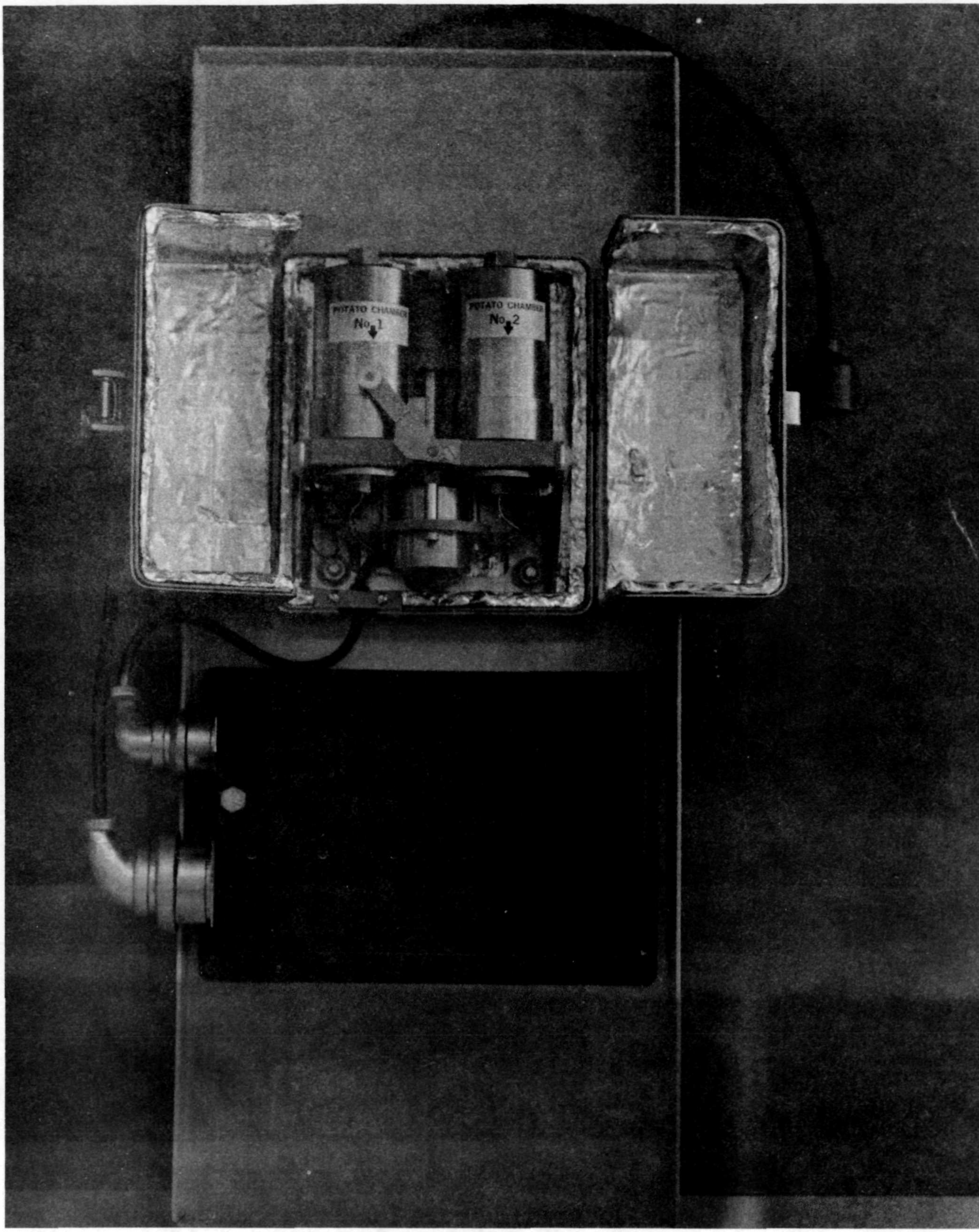


Figure 1. Flight Hardware Configuration of Potato Respirometry Experiment. Upper Unit is Life Support System Showing Thermal and Protective Cover Doors Open. Lower Unit Contains Electronics and Data Storage

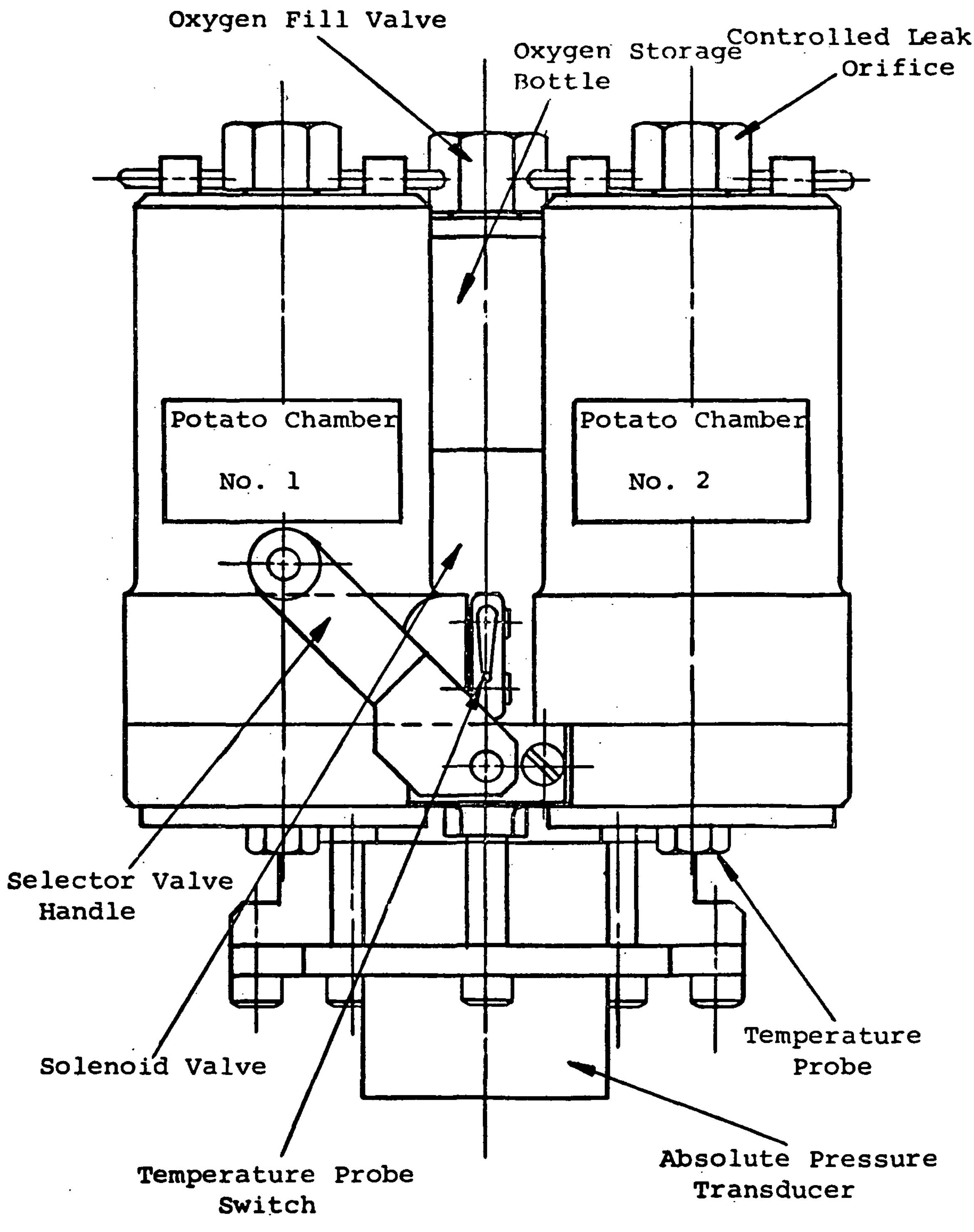


Figure 2. Identification of Major Life Support System Subassemblies

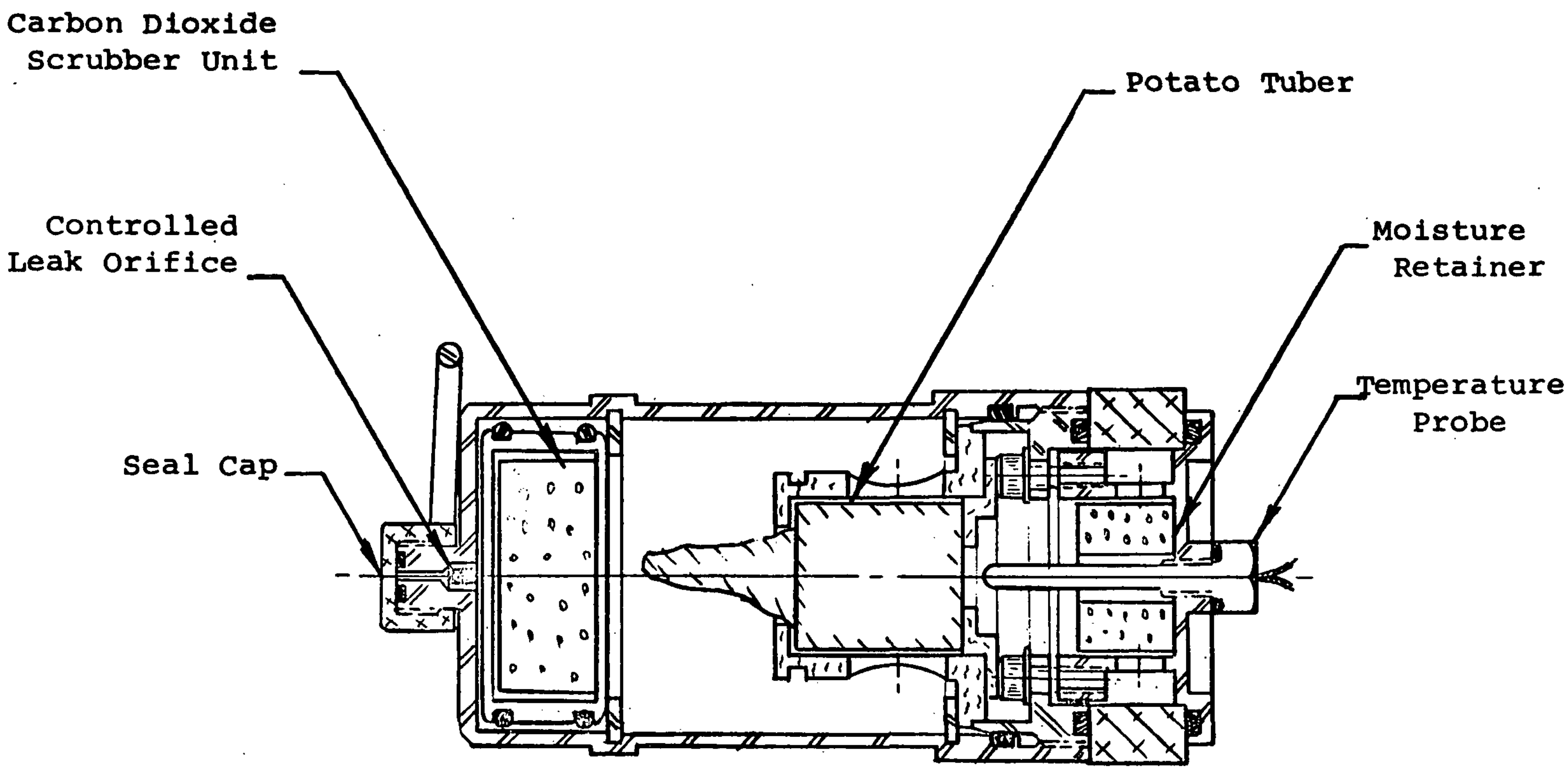


Figure 3. Cutaway View of Potato Chamber

is supported by a Teflon holder. Humidity is provided by a saturated sponge. All light is excluded to limit metabolism to the non-photosynthetic mode only. The carbon dioxide produced is removed from the environment by a potassium hydroxide impregnated sponge. This sponge is enclosed in an open ended Teflon cylinder, Figure 3, with the ends closed by a semipermeable membrane. Scrubber reaction products are thus prevented from dripping on and damaging the specimen. In order to minimize the effects of corrosion on chamber walls and subsequent potential damage of the leak tight O-ring seal, the cover is made of stainless steel. Figure 4 is a photograph of a semi-disassembled PRU.

Temperature data is acquired and temperature control is achieved by the platinum resistance probes located in the base of each potato chamber. The probes have a resistance of 470 ohms at room temperature and a temperature range of $-100^{\circ}C$ to $+204^{\circ}C$. Calibration accuracy is approximately $\pm 0.007^{\circ}C$, (see Section 6.5) and is NBS traceable. Data is acquired from the probe located in the base of the active chamber while control is achieved via the probe in the standby chamber. Probe functions are reversed via the temperature probe switch when the back-up specimen is activated. Heat is provided on demand by a four watt resistance type heater strip cemented to the back of the manifold block.

Oxygen is contained in the 16.39cc storage bottle at 8.53 meters Hg. This supply is sufficient for a 100-day mission at nominal specimen oxygen consumption rates. To meet NASA safety requirements, the bottle is designed for and has passed a differential burst pressure test at 34.12 meters Hg. Five micron

Figure 4 Flight Configuration
Life Support System, Partially Disassembled

filters are provided at the oxygen bottle fill valve and solenoid valve input to minimize contamination. The solenoid valve permits resupply of oxygen to the specimen as required. This valve, manufactured by Wright Components, Inc., operates on 22.0 - 32.0 VDC at one watt and has a nominal leak rate of 2 x 10^{-8} ml/sec. The controlled flow orifice limits the charging rate of the potato chamber to a value to which the absolute pressure transducer can respond without pressure overshoot. Figure 5 shows these components and the gas flow path.

The selector valve serves several functions. It directs the flow of oxygen to one or the other of the potato chambers, and simultaneously exposes the absolute pressure transducer sensing port to the environment of the active chamber. The selector valve handle location is shown in Figure 2. The position of the handle indicates the active chamber. A design feature inhibits inadvertent removal of the active potato chamber cover. In addition, moving the selector valve handle from one potato chamber to the other activates an electronic switch located directly behind the handle. Activation of the switch directs the output of the proper temperature probe to the data channel and reverses the control and measurement functions of the two probes. The manifold block provides mechanical support and includes pneumatic interconnections of components.

3.1 Servo Pressure Sensor

The key component in the respirometer system is the servo pressure sensor developed by the Kistler Instrument Company*

*Patents granted and pending

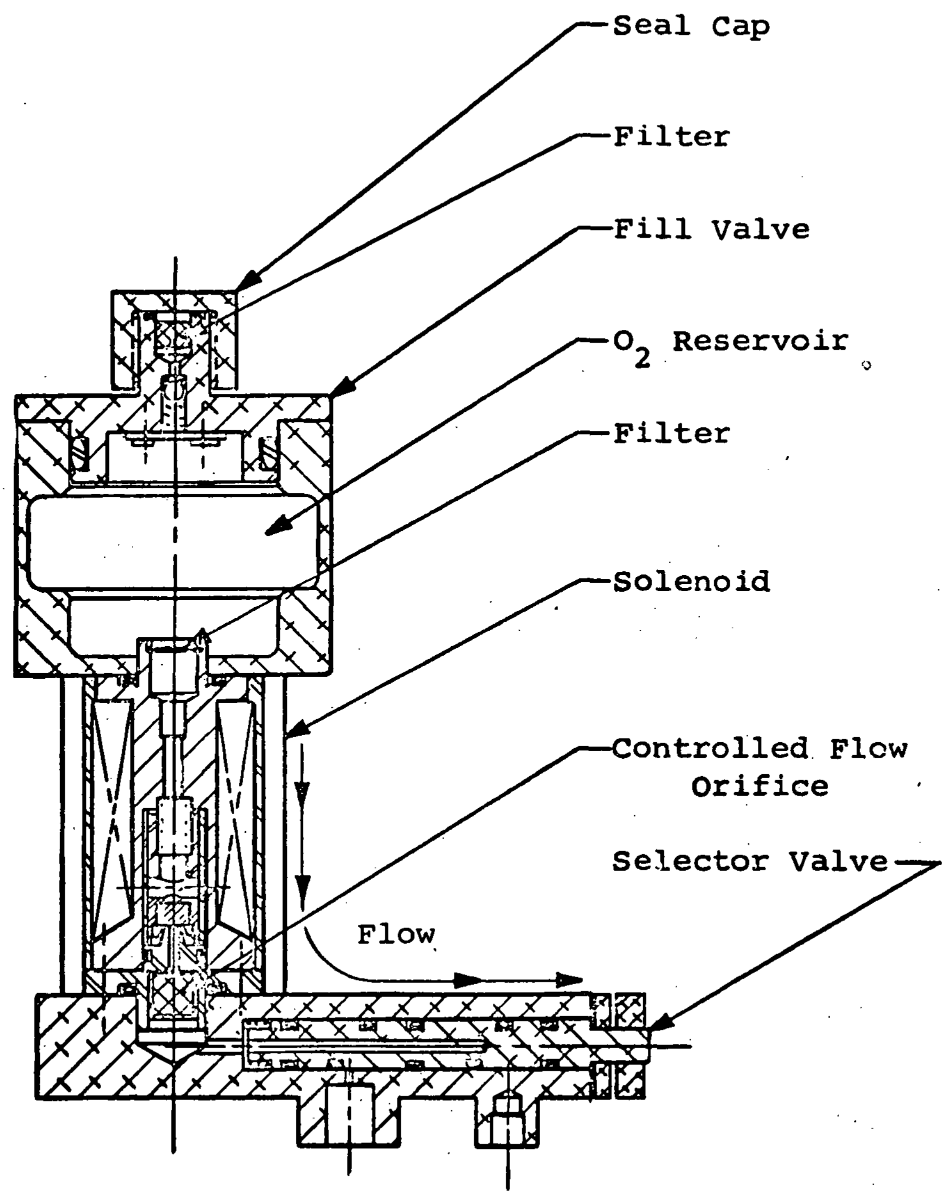


Figure 5. Potato Respirometer Gas Flow Path

This force-balance type instrument has met the requirements of extreme accuracy, high resolution, small size and signal output. This transducer acquires pressure data and provides the signals for absolute pressure control.

This instrument is of the force-balance or closed loop type. A pressure input produces a force which deflects a movable assembly. The motion is detected and a counteracting force is generated by a current passing through a coil which is positioned in the field of a permanent magnet. The amount of electrical current necessary to restore the sensor toward its null position is proportional to, and a measure of, the applied pressure. A voltage output is produced by passing the current through a precision resistor in series with the force-balance coil. By making the amplifier gain and pickoff sensitivity very high, extremely small deflections of the novable assembly are sufficient to produce full scale output. The electrical force feedback is thus equivalent to a very stiff spring and the high constrainment results in excellent repeatability and low hysteresis. The pressure transducer contains a mechanical assembly consisting of an open bellows-bending beam-reference bellows arrangement such as to yield a torque about a single sensitive axis when pressure is applied to the open bellows. The appropriate electronics consists of a detection device to sense angular deflection and a torquer which restores the mechanical assembly toward its null position.

Special bias and amplification networks were designed to provide a 0 to 5 volt signal output for the limited pressure range of 755 to 770 mm Hg. Since the sensor is in its neutral assembled position at one atmosphere or 760 mm Hg, minimal current is required through the force-balance coil resulting in low power drain.

Table II shows the performance and environmental specifications for the Kistler Instrument Company's Model 311 Servo Pressure Sensors. Performance is specified over the 760 mm full scale range and the results stated as a percent of full scale output (FSO).

3.2 Respirometer Electronics

The DSU provides all the electronic support for the Flight Hardware. Figure 6 is a block diagram of the DSU. An interconnecting cable between the DSU and PRU provides power to the absolute pressure transducer, the solenoid valve, and the PRU heating unit. The PRU transducer outputs are returned to the DSU through the interconnecting cable. The DSU is hermetically sealed at 950.0 $\pm$ 48.0 mm Hg with a 90% nitrogen and 10% helium gas mixture. This special environment inhibits combustion and prevents any seepage of spacecraft contaminants into the DSU. A one-way fill valve required for pressurization is located on the side of the housing. The DSU has three modes of operation: data acquisition, data dump, and data clear. Power is provided from the CM at 25 to 30 VDC and is regulated by an internal power supply.

Several of the major DSU elements are functionally described as follows:

TABLE II

Performance Data of Kistler Model 311

Servo Pressure Sensors

Pressure Range	0 - 762 mm Hg (absolute or differential)
Sensitivity (nominal)	5V FSO*
Linearity	0.06% FSO
Hysteresis	0.02% FSO
Repeatability	0.02% FSO
Null stability	0.05% FSO
Resolution (noise level)	0.001% FSO
Temperature zero shift	0.002% FSO
Temperature sensitivity shift	0.001% / °C
Linear acceleration sensitivity	0.03% FSO/g
Resonant frequency	100 Hz
Power supply variation:	
Null	0.01% FSO/V Max.
Sensitivity	0.05% / V Max.
Temperature range (operating)	-54°C to +80°C
Vibration	10g, 15 to 500 Hz
Shock	20g 10 ms pulse
Size	4.12 cm dia. x 4.82 cm long overall
Weight	225 grams

*Absolute mode output is +5V at vacuum
0V at 760 mm Hg
-5V at 1520 mm Hg

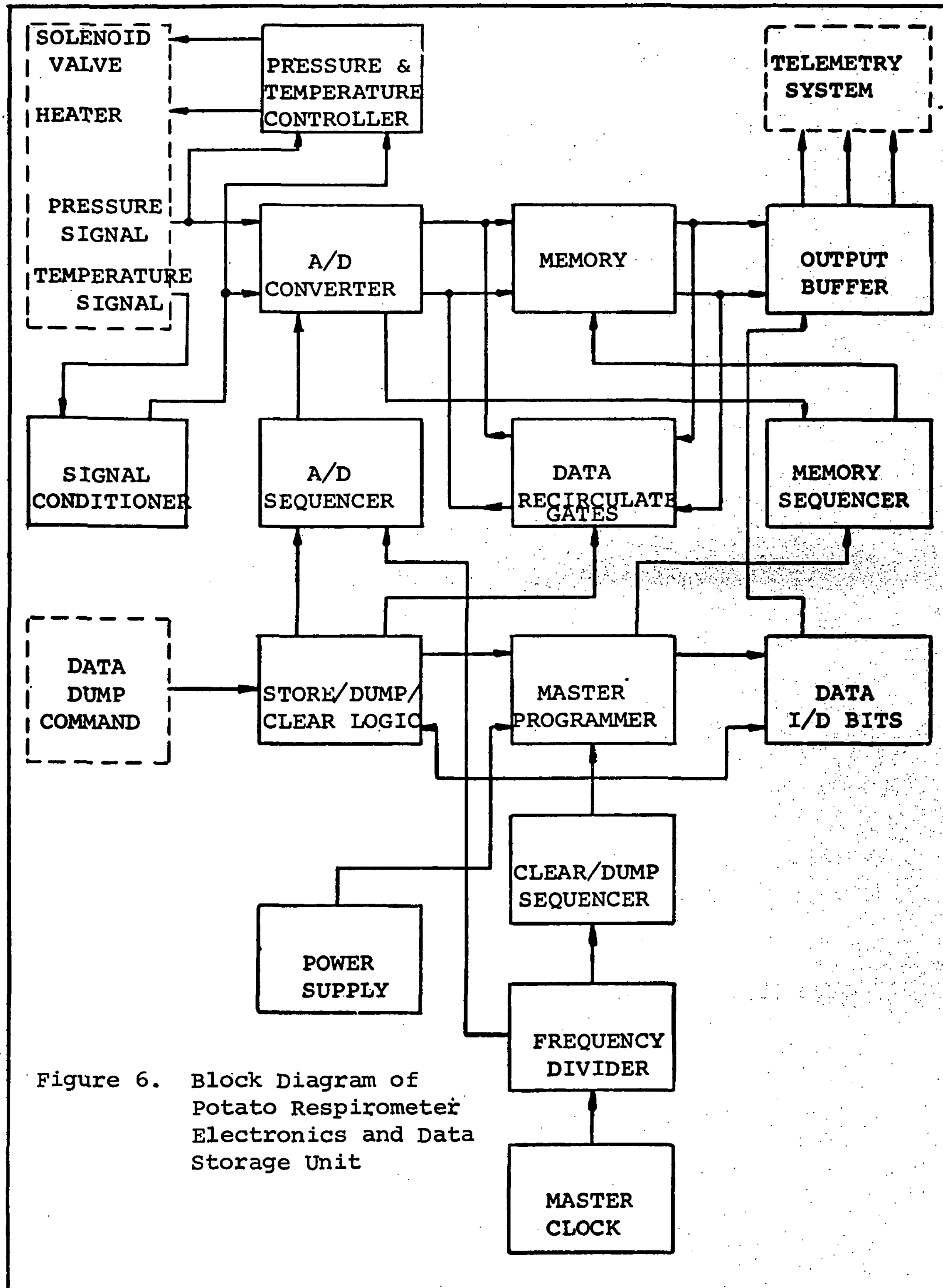


Figure 6. Block Diagram of Potato Respirometer Electronics and Data Storage Unit

3.2.1 Signal Conditioner

The temperature sensing element is part of a bridge circuit whose output is 0 - 5 mvdc. The output of the bridge is amplified by a factor of 1000 providing a 0 - 5 vdc signal to the temperature controller and the A/D converter.

3.2.2 A/D Converter

The analog temperature and pressure signals from the PRU are converted into eight bit binary words that are then conditioned and stored in the memory.

3.2.3 Memory

The memory element consists of shift registers that store up to 300 parallel words that represent five hours of temperature and pressure data. The memory sequencer provides pulses required to store data. Upon data dump command, data are recirculated to the memory input and presented to the output buffer twice.

3.2.4 Output Buffer

The output buffer ensures proper voltage levels corresponding to a binary 1 and a binary 0 at the DSU telemetry output. The DSU output consists of three eight bit words representing temperature and pressure measurements and data identification.

3.2.5 Master Programmer

The master programmer controls and sequences system functions throughout the DSU. The master clock generates the 1.0 kHz time base that is divided and distributed to the various subsystems. Appropriate logic gates control, store, dump, and clear functions. Three data identification bits are generated that identify: (a) valid and invalid data, (b) an odd and even measurement and (c) the first and second reading of the memory element.

3.2.6 Temperature and Pressure Controller

The analog temperature signal is compared to a pre-set reference voltage representing 23.3°C. When the temperature falls below the reference, full power is applied to the heater until the temperature rises slightly above the correct value. The error is determined by the hysteresis of the voltage comparator.

Pressure in the potato chamber is controlled by a hysteresis voltage comparator. When the pressure falls to 758.0 ± 0.1 mm Hg, the solenoid valve is energized until the pressure reaches 762.0 ± 0.1 mm Hg.

3.2.7 Power Supply

The CM power bus is regulated and converted to voltages required by the experiment subsystems. EMI filters and short circuit protection are provided.

3.2.8 Respirometer Operation

Figure 7 is a functional schematic of the respirometer and shows all spacecraft interfaces. As the potato breathes, oxygen is consumed and carbon dioxide is formed that is removed by passive "scrubbers" located in the chamber above the specimen. The 5.50 cc specimen consumes oxygen at a nominal rate of 0.06 ml/hr. When the pressure drops to 758.0 $\pm$ 0.1 mm Hg, the chamber pressure is increased to 762.0 $\pm$ 0.1 mm Hg by the solenoid valve opening and admitting fresh oxygen from the storage bottle. Refill rates range from approximately two seconds to three minutes over an oxygen bottle pressure range of 850 cm Hg to 100 cm Hg. The free volume of the potato chamber is 83.5 cc. The solenoid valve operation is controlled by the absolute pressure transducer and its output is continuously sampled and compared to a stored voltage level reference in the control logic system. The flight configured electronics operates as follows: In the data acquisition mode, the store logic circuitry enables the A/D sequencer once per minute for sampling temperature and pressure. The A/D sequencer provides pulses required for conversion of the analog signals into parallel eight bit binary words. After the conversion has been completed, the A/D converter pulses the memory sequencer, permitting the storage of the data in the memory. A data identification word is generated and stored in the master programmer corresponding to each data point. When the memory is full, the memory sequencer is inhibited so that stored data are not lost. Upon data dump command, appropriate logic gates disenable the A/D sequencer and enable the data recirculate gates. The dump sequencer is enabled

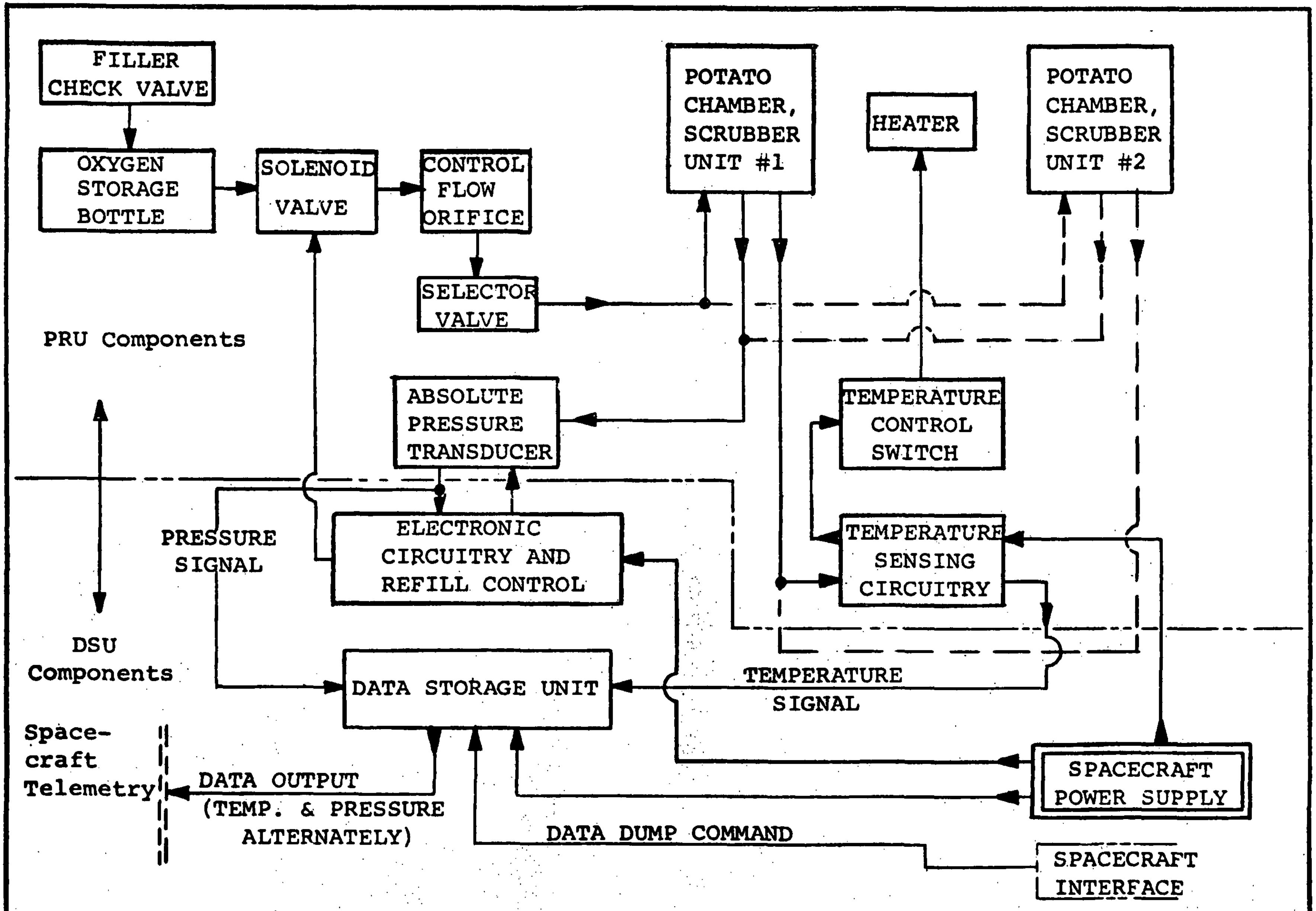


Figure 7. Block Diagram of Potato Respirometer

107 msec after the dump command is received. The contents of the memory are recirculated to the memory input and presented to the output buffer twice. If the memory is not full, any empty memory location will be shifted at one word/msec until data are present at the memory output. Data are presented to the output buffer at one word per msec. This rate minimizes the CM PCM samples that occur while the data shift is in process. If the DSU output is sampled while the data shift is in process, the datum bit that is present will indicate invalid data. After the memory has been presented twice, the memory is cleared in preparation for new data and the system is returned to the data acquisition mode.

4.0 RESPIROMETER LABORATORY FACILITY

4.1 General

In order to meet the revised objectives of the potato respiration program, a complete laboratory facility was designed to permit thorough evaluation of potato sprout oxidative metabolism. Particular emphasis was placed on control of ambient temperature changes in order to eliminate, in so far as possible, temperature induced rhythmic artifacts. A complete data acquisition system and a system for calibration of the pressure transducers in assembled respirometers under control temperature conditions were also designed and built.

The laboratory facility consists of four major units:

1. Controlled temperature water bath
2. Waterproof respirometer containers
3. Pneumatic system for transducer calibration
4. Control electronics and data acquisition system

4.2 Controlled Temperature Water Bath

The controlled temperature bath was designed around a large double walled fiberglass tank (1.9 x .45 x .51 meters, 120 gallons) with a 3950 BTU refrigeration unit. Water was recirculated by the cooler at a rate of 4500 gallons per hour.

The primary objective of the system was to control respirometer temperature as accurately as possible in a room temperature ambient which varied by as much as $\pm$ 3.0°C during a 24 hour period. In addition, water temperature was to be maintained at 20°C, 5°C cooler than average room ambient.

Figure 8 is a photograph of the water bath facility. The top is covered with a 7.5 cm thick sheet of polyurethane foam. Figure 9 is a schematic of respirometer location, temperature regulation equipment and water flow baffle arrangement. Baffle arrangement was determined by trial and error to provide uniform (through water bath cross section) but semi-turbulent flow. The high pumping rate provided for complete recirculation of the water every 1.5 minutes. Turbulent mixing occurred primarily due to the circular shape of the water proof containers and was identified by adding drops of blue dye to the water at selected points.

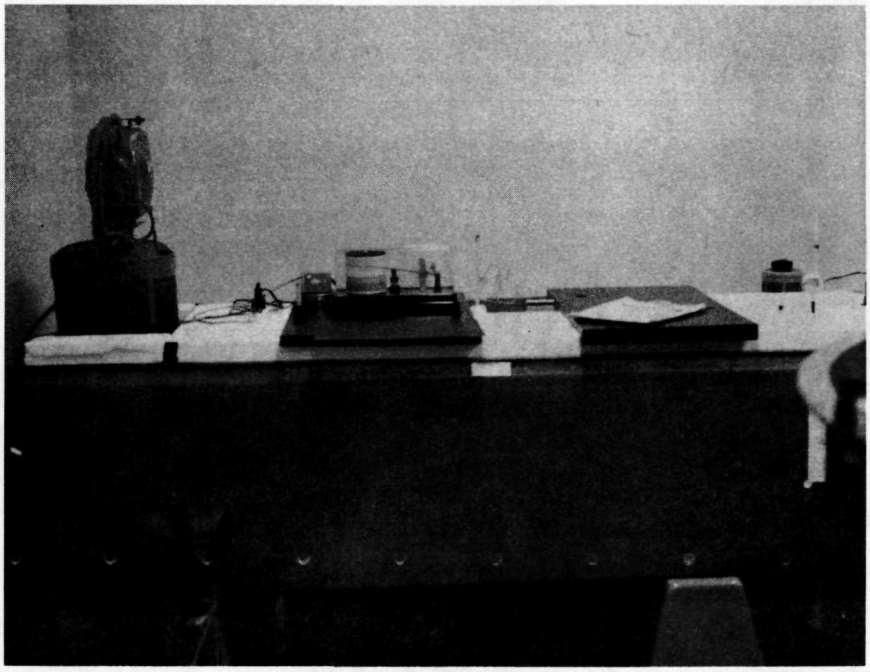
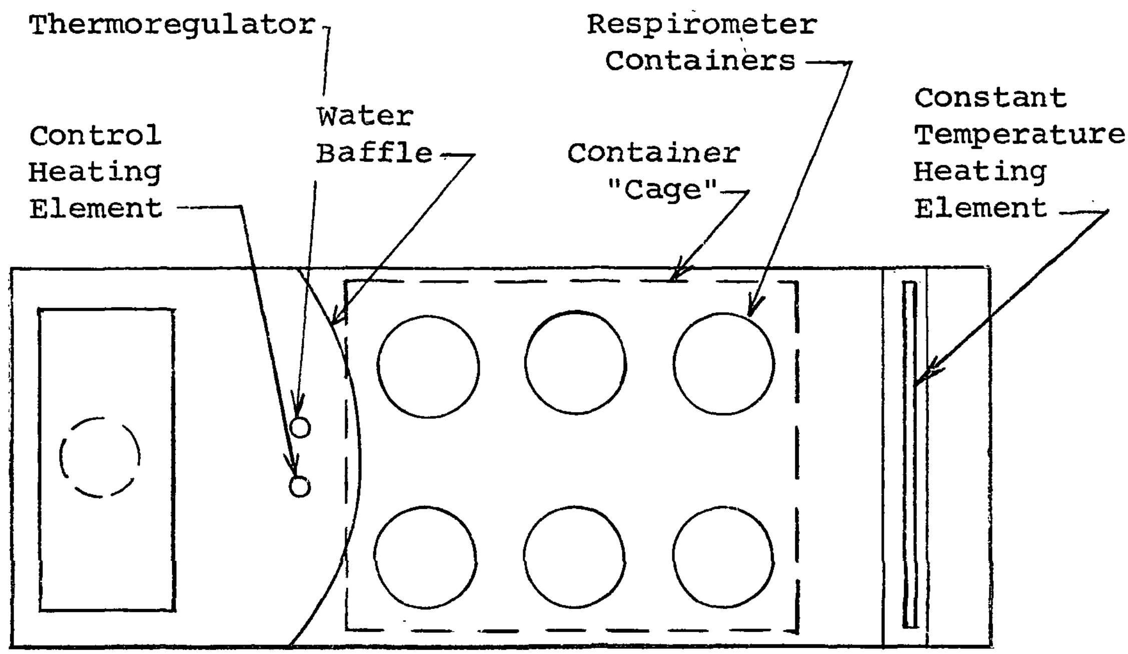


Figure 8. Photograph, Water Bath Facility



Water Bath: Top View

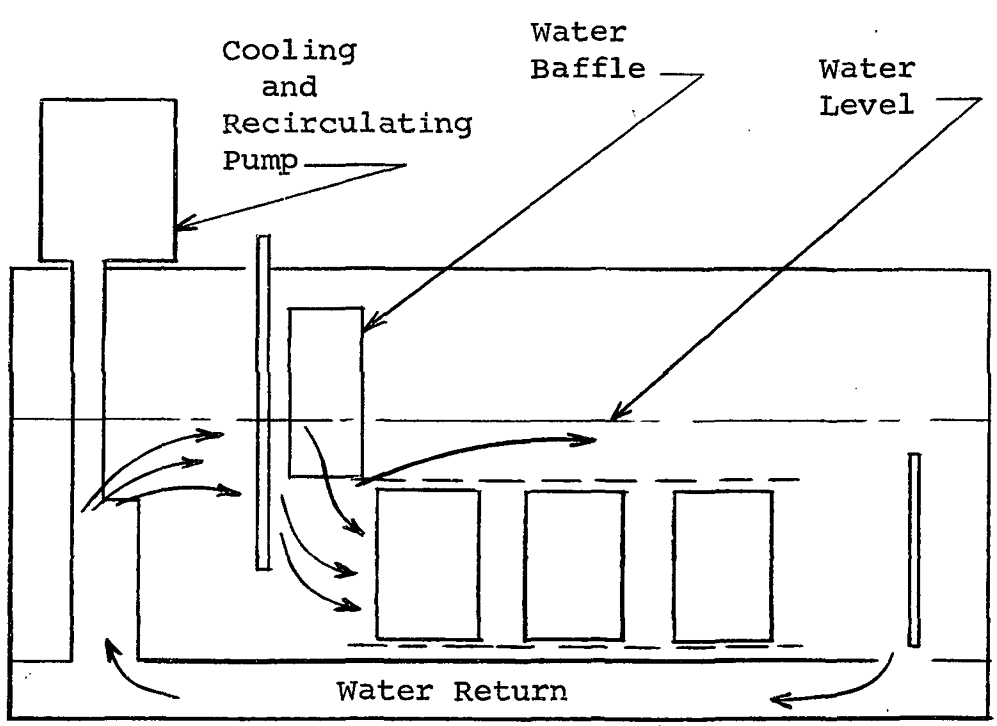


Figure 9: Water Bath: Side View

A constant voltage transformer supplies power to the constant cooling unit located at one end of the tank and a constant heating unit located at the opposite end. The temperature of the water bath is controlled by a thermoregulator, control heating unit, and a zero voltage switch. The thermoregulator provides a contact closure when the temperature of the bath is above the preset value and an open circuit when the temperature falls below the preset value. This switching action is sensed by the zero voltage switch which operates the control heating unit to maintain constant temperature.

Performance of the water bath was monitored with precision (0.01°C) laboratory differential thermometers immersed in the bath and by the temperature probes mounted in each respirometer. Since each pressure transducer dissipated approximately 1.0 watt of power during operation, respirometer equilibrium temperature was slightly above water bath ambient. In addition, differences in thermal conductivity properties of each container (slight differences in respirometer position and in copper "sponge" configuration) resulted in different equilibrium temperatures for each respirometer. Representative respirometer temperatures are given below:

TABLE III

Internal Respirometer Temperatures
With Water Bath at 20.00°C

Unit	1	3	5	7	8	9
Internal Temperature (°C)	21.647	21.634	21.357	21.291	22.106	21.398

Constancy of water bath temperature has been studied for periods of 2 - 3 days over a three month period. Maximum observed temperature fluctuation over a 24 hour period was $\pm$ 0.0075°C. Results were obtained using the internal respirometer temperature probes. Data was automatically recorded at 5 minute intervals 24 hours a day using the digital printer.

4.3 Water Proof Respirometer Containers

In order to provide the fine temperature controls required by this experiment, fail-safe waterproof containers were required for each respirometer to permit immersion in a controlled temperature bath. Since pressure transducer calibration also had to be performed under temperature controlled conditions, the container had to permit access to each individual unit via metal tubing.

Figure 10 is a photograph of the containers and the calibration pneumatic system. The containers are metal cans with a ring-clamp seal for a cover. Prior to use, each cover was liberally coated with silicone grease to assure a water tight seal. Electrical connections to the respirometer were made using a standard, NASA approved, multi-pin waterproof electrical connector. Pneumatic connection was accomplished by using standard waterproof Swagelock fittings.

Respirometers are installed in the containers and the remaining free space filled with commercially available copper "sponges" to improve heat exchange between the respirometer and the container.

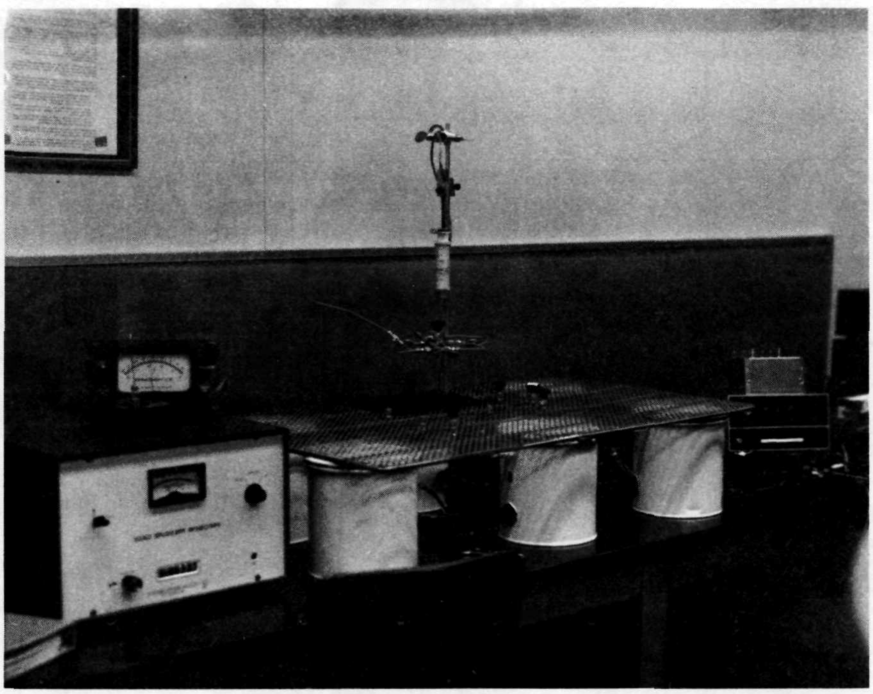


Figure 10. Respirometer Water Proof Containers and Tubing for Pressure Transducer Calibration

As an added safety feature water detectors are located in the bottom of each waterproof container. These are necessary since water immersion could produce irreparable damage to the pressure transducers. The detectors consist of two metallized strips on a printed circuit board. If a water leak occurs, the strips are shorted and a warning light will be activated. Electrical connection to the sensors is accomplished through the respirometer waterproof electrical connector.

4.4 Pneumatic System for Pressure Transducer Calibration

Three factors are essential in calibration of the respirometer pressure transducers:

1. Constant temperature
2. Ability to vary temperature (to determine temperature sensitivity)
3. Ability to set a known pressure precisely.

The system for calibration is shown schematically in Figure 11. Completely assembled respirometers were immersed in the water bath. Each unit was connected to the pressure manifold by stainless steel 1/16" I.D. tubing using Swagelock connectors. Each unit could be isolated from the manifold via a valve. Connection to the respirometer inside the can was accomplished by heavy walled tubing 4.5 inches long fitted over the orifice on top of the specimen chamber cover. The pressure manifold terminated at the 100 cc syringe and at the Texas Instruments Bourdon Tube Pressure Standard. A micrometer type attachment to the syringe handle permitted precise, repeatable setting of the desired pressures.

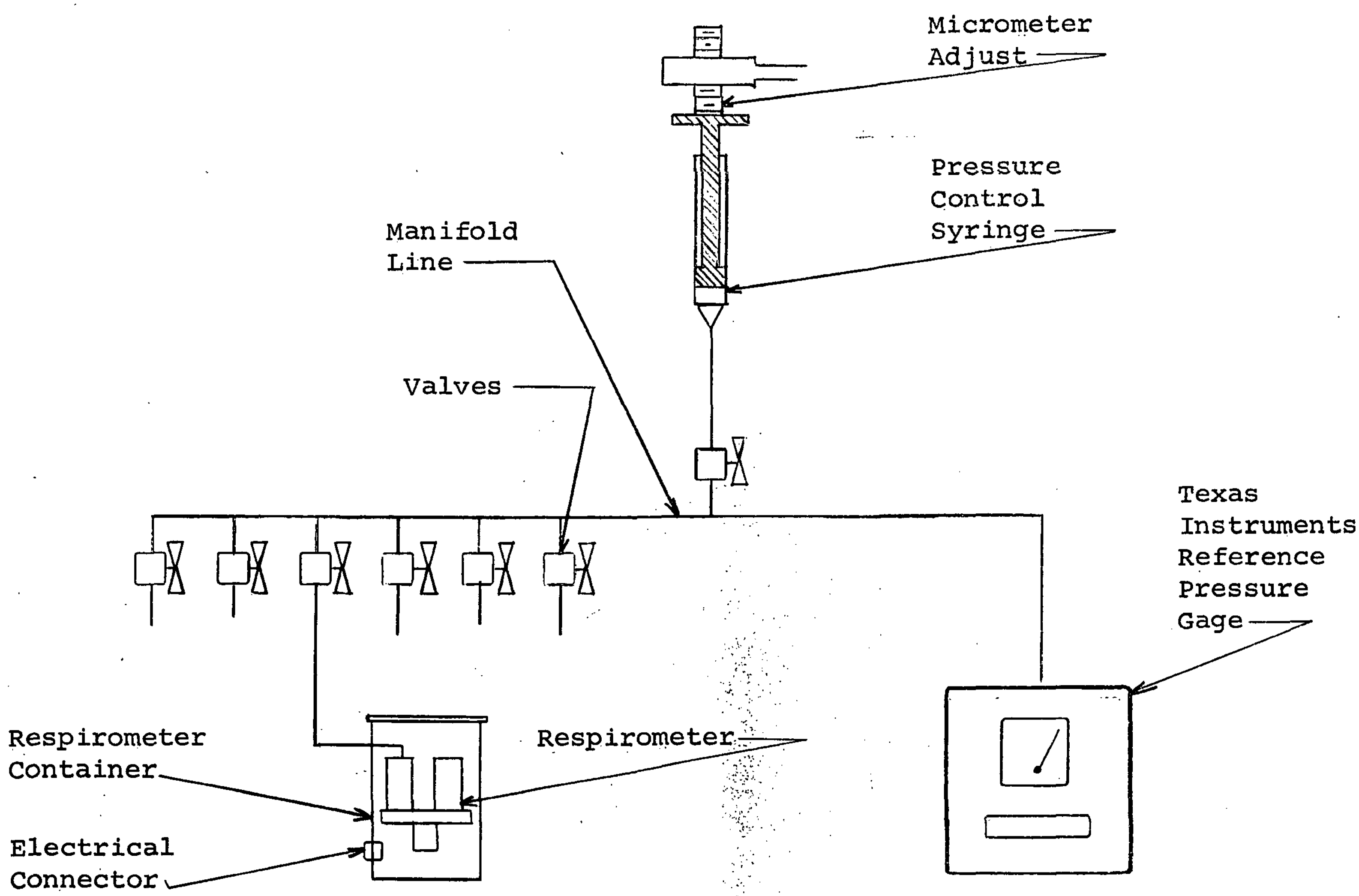


Figure 11. Schematic: Pressure Transducer Calibration System

All water bath electronics and the TI gauge were operated off a heavy duty 1% line voltage regulator. Respirometer voltages were regulated to ± 0.01 volts.

5.0 CONTROL ELECTRONICS AND DATA ACQUISITION SYSTEM

5.1 Introduction

The system is designed to replace the data storage unit portion of the SO61 flight hardware. Temperature and pressure data are sampled at preset time intervals and recorded on paper tape and on magnetic tape. The system consists of a printed circuit card rack, a scanner, a digital voltmeter, a digital printer, a tape recorder interface unit, and a digital tape recorder.

5.2 System Operation

The block diagram, Figure 12, shows the operation and interconnection of the system.

The card rack contains 6 identical circuit cards to condition the data from the potato respirometer. A cable from the card rack carries the temperature and pressure data to the scanner.

When the clock in the scanner reaches the set point a sample command is given to the digital voltmeter. This causes the DVM leads to be switched to the first channel and the DVM takes a measurement of the voltage on that line.

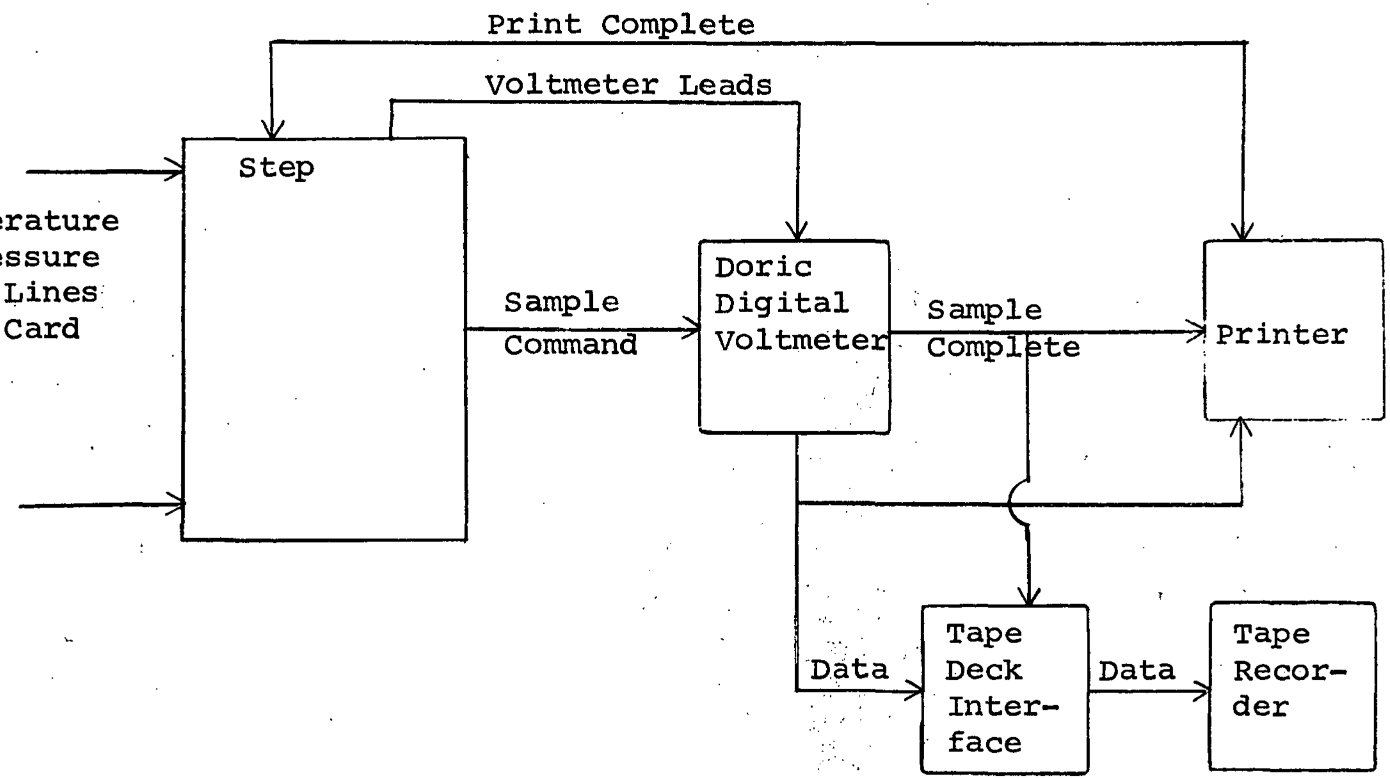


Figure 12. Block Diagram: Data Acquisition System for Potato Respirometers

The DVM converts the voltage level into binary coded decimal format and commands the printer and the tape recorder interface unit. The tape recorder interface unit converts the data into IBM compatible format and commands the tape recorder to record the data. At the same time, the digital printer prints the data on paper tape and commands the scanner to switch to the next channel and sample. This process continues until all selected channels are sampled.

5.3 Component Function

5.3.1 Printed Circuit Card Rack

The card rack contains 6 identical printed circuit cards, one for each respirometer. Each card has an amplifier for the temperature signal and conditioners for both temperature and pressure signals. A controller maintains the pressure in the respirometer between 758 and 762 mm Hg ± 24 VDC and ± 15 VDC must be supplied from high quality power supplies.

5.3.2 Scanner

The scanner samples temperature and pressure data by switching the DVM leads to each channel in succession and commanding the DVM to sample the voltage. This occurs automatically at a preset time interval. A push-button switch is also provided for manual operation. The scanner is capable of sampling 17 channels of data. The switches on the front panel determine whether that particular channel is to be included in the data sample. A switch in the up position includes that channel in the data sample.

On the rear panel are located the data input terminals, power terminals, and command signal terminals to the other system components.

5.3.3 Digital Voltmeter

Both pressure and temperature output voltages are measured by a Doric Model DS-100 Integrating Digital Voltmeter. The DVM measures the voltage present at its input leads and converts the voltage into binary coded decimal format when commanded by the scanner. When the sample is complete the data is presented to the digital printer and the tape deck interface. A print and record command is also given.

5.3.4 Tape Deck Interface

The tape deck interface unit converts the data from binary coded decimal format to IBM compatible format for storage on magnetic tape.

5.3.5 Tape Recorder

The recorder is an incremental digital magnetic tape unit manufactured by Digi-Data Corporation, Model 1420. Data format was designed to be compatible with the NASA Univac Model 1108 computer. Data format is 10 character binary coded decimal (BCD) with six bits per word. The character assignment is:

a. 2 bit - channel identification

b. 1 bit - decimal point location

c. 1 bit - sign

d. 5 bit - data

e. 1 bit - word end

5.3.6 Printer

The digital printer, Hewlett-Packard Model 5055A, prints the data on paper tape in analog format. After the print is complete a signal is sent to the scanner to step to the next data channel.

6.0 RESPIROMETER PERFORMANCE

6.1 Introduction

Evaluation of respirometer performance was accomplished both in the newly developed laboratory and during the Special Test Program requested by NASA and performed at the University of California, Davis. All major respirometer performance characteristics pertinent to oxidative metabolism measurement were studied including:

a. Determination of the total absence of instrumental rhythmic artifacts.

b. CO_2 scrubber performance

c. Life support chamber leak rate

d. Pressure transducer calibration

e. Temperature transducer calibration

Items a and b were analyzed during the Special Test Program and are fully discussed in reference (3). A brief review of these results is given below. Samples of potato sprouts are shown in Figure 13. Figure 14 shows four loaded respirometers with life support chamber cap removed.

6.2 Instrumental Rhythmic Artifact

Measurement of biologic rhythms requires that all instrumental or environmentally induced rhythms be eliminated. There are two reasons for this: (1) a rhythm external to the specimen under study may entrain or otherwise influence specimen behavior and (2) the artificial rhythm may be interpreted as specimen behavior. The problem of rhythmic artifacts is of particular concern for the phenomena under consideration here because of the very small perturbation studied. The extremely sensitive equipment required for pressure and temperature measurement is also sensitive to potential rhythmic environmental factors. Three potential sources of rhythmic artifacts exist in this respirometer: rhythmic ambient temperature, rhythmic gas leaks and possible rhythmic scrubbing action in the CO_2 scrubber. The most serious potential rhythmic artifact source is room temperature changes. Since respirometric data is obtained from measurement of pressure and temperature, care must be taken to assure that the respirometers are completely isolated from room temperature changes. The problem is compounded by the fact that both controlled temperature buildings and the atmosphere exhibit 24 hour periodic fluctuations. Care also must be taken to assure that system electronics, especially signal processing equipment, is not sensitive to ambient temperature changes.

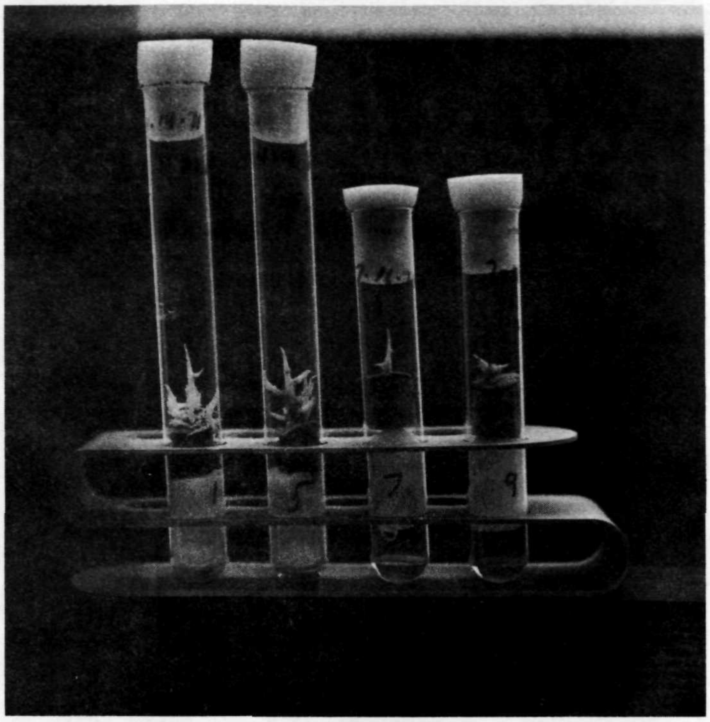


Figure 13. Samples of Potato Sprouts in Storage Tubes

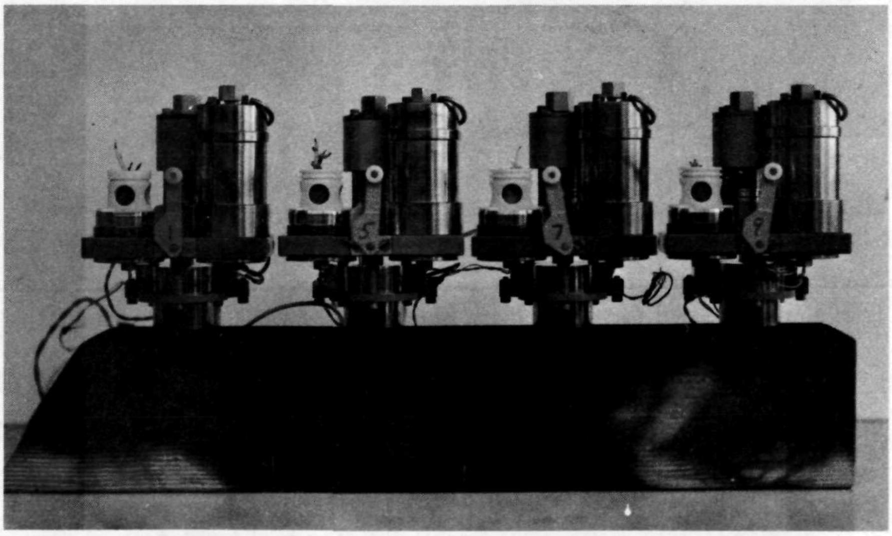


Figure 14. Specimens Installed in Respirometers with Life Support Chamber Cover Removed

Both empty respirometers and a respirometer containing a yeast colony have been studied. No unexplainable rhythms of any kind were detected.

Empty units operated in temperature controlled water baths exhibited no detectable rhythmic behavior. Temperature control was $\pm 0.01^{\circ}C$ at UCD and $\pm 0.005^{\circ}C$ at COMAR laboratories. The yeast test, also performed at UCD, is a well known technique for evaluation of respirometer performance. An active yeast colony consumes oxygen and gives off carbon dioxide as do plants growing in a non-photosynthetic mode. The oxygen consumption rate of a colony can be adjusted simply by changing its size. In addition, yeast is known to be arrhythmic because of its totally random organization. A yeast colony supported by a glucose, agar-agar medium and exhibiting an oxygen consumption rate of approximately 100 microliters per hour was studied. No rhythmic behavior was observed.

Leak tests were performed on all six empty units. Leak rates observed were in the range of 1×10^{-7} to 5×10^{09} cc/sec. No rhythmic behavior was observed in empty units as noted above.

The lack of rhythmic behavior in the yeast colony study indicates that no rhythmic scrubbing action occurs.

6.3 Carbon Dioxide Scrubber Performance

The carbon dioxide scrubber has been thoroughly evaluated. Potato respiration studies lasting 120 days have been successfully performed with no indication of scrubber saturation.

The two primary concerns about the scrubber have been saturation and scrubbing lag. The latter phenomena implies that a certain critical partial pressure of CO_2 would be required before scrubbing was initiated. This would result in a rhythmic pumping effect as the CO_2 partial pressure increased to the level required for scrubbing and would also result in exposure of the specimen to unacceptably high CO_2 levels. This phenomena was not observed except at the initiation of an experiment and was related to the requirement that the scrubber be slightly moist for most efficient scrubbing. This lag lasted 1 - 3 hours.

In addition to the long term tests, two others were performed. A very large yeast colony was placed in the respirometer. The oxygen consumption rate, and the CO_2 evolution rate was 50 times that of a normal potato. Oxygen refills occurred every eight minutes. No scrubbing lag was observed and although the amount of CO_2 equivalent to a 40 day mission was generated in less than 24 hours, no saturation occurred.

Finally, two alternate types of CO_2 scrubbers were used as a check of the KOH system. Barium Hydroxide and Baralyme* were evaluated. Small changes in oxygen consumption rates were noted. However, there was no means of determining if this change was simply due to increased potato metabolic rate or not.

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*Collins Baralyme Granules
Warren E. Collins, Inc.
555 Huntington Avenue
Boston, MA

6.4 Life Support System Gas Leaks

A leak tight respirometer is important for several reasons:

a. A leak would indicate an unrealistically high oxygen consumption rate.

b. A constant leak superimposed on the potato consumption rate would tend to obscure the rhythmic behavior, if present.

c. It is critical in a study related to metabolism that all environmental variables remain constant. A sizeable gas leak would, eventually, reduce the partial pressures of other atmospheric gases and, therefore, result in a higher than normal oxygen partial pressure.

Many leak tests were conducted during the respirometer development program. Prior to and during pressure transducer calibration all six empty units were maintained in the constant temperature bath for up to four weeks. Leak rates varying from 1×10^{-7} to 5×10^{-9} cc/sec were measured. The design goal was 1×10^{-7} cc/sec or 1/10 the anticipated niminal oxygen consumption rate for a specimen.

6.5 Temperature Transducer Calibration

Temperature transducer calibration was performed at Leeds and Northrup Corporation Standards Laboratory. Calibration included the COMAR supplied amplifiers and digital voltmeter. Calibration was performed by placing them in a constant temperature oil bath in close proximity to a standard platinum resistance thermometer. Reference temperature was recorded to an accuracy of $\pm$ 0.0001°C and voltmeter output to $\pm$ 0.0001 volts.

The raw calibration data was analyzed and a calibration table prepared by Dennis Johnston, NASA, Houston.

Data analysis was performed using the polynomial regression technique. The basic computer program used is available at the Scientific Computing Branch, Computation and Analysis Division, NASA/MSC, Houston, Texas, and is identified as NASA Routine CMSCC089.

The temperature calibration range of the respirometer temperature probes was 18.01°C to 26.00°C. A calibration table has been prepared in increments of 0.01°C. The complete calibration report is on file at COMAR Corporation. Table IV gives an indication of the accuracy of the calibration curve. Probe numbers refer to respirometer number and are identified further by pressure transducer serial number. Probes 2, 4 and 6 were not used because of noisy amplifiers. These were prototype units and were not intended for use in flight hardware.

The best fit polynomial was, in all cases, a second degree polynomial of the form:

$$V = a_o + a_1T + a_2T^2$$

TABLE IV

Summary of Calibration Results for Respirometer Temperature Probes

Probe No.	Sample Standard Deviation (°C)	Approximate 95% Confidence Level (°C)
1	0.0018	0.0036
3	0.0020	0.0040
5	0.0023	0.0046
7	0.0017	0.0034
8	0.0028	0.0056
9	0.0015	0.0030

6.6 Pressure Transducer Calibration

Pressure transducer calibration was performed using completely assembled respirometers under controlled temperature conditions.

The pressure reference was a Texas Instruments Company 0 - 770 mm bourdon tube absolute pressure gauge (Model #144, SN4245). This gauge is commonly used as an absolute pressure standard. This unit had been calibrated by the manufacturer to an accuracy of ± 0.004 mm Hg and a 10,000 point computer interpolation provided. The digital output of the gauge could be read to ± 1 count or ± 0.004 mm Hg.

Calibration was performed by varying the pressure in 1.0 mm steps in the range 755 to 765 mm Hg as measured by the pressure standard. Pressure was varied by adjusting the volume of the calibrated (1 cc per division) syringe shown in Figure 11. Adjustment of pressure to an accuracy of ± 0.004 mm Hg was readily achieved by this technique. Each respirometer was calibrated in the pressure increasing and pressure decreasing modes. (P^+, P^-, P^+) at temperatures of 20.00, 21.68 and 24.00°C. Transducer output was monitored by a digital voltmeter accurate to ±0.001 volts and the result recorded by a digital printer. The output was recorded at each pressure point after allowing five minutes for pressure and temperature stabilization and if three successive readings taken five minutes apart were the same. Some difficulty occurred in that the syringe was not immersed in the water bath and room temperature air was introduced into the respirometer. Over the entire calibration program, the maximum internal temperature change recorded was 0.050°C. Based on the measured temperature sensitivity of the transducers, the maximum temperature induced output error at any pressure for units 5, 7, 8 and 9 was ± 0.0012 volts. Units 1 and 3 exhibited a substantially larger temperature sensitivity.

Table V lists the temperature sensitivity for each unit as measured by change in voltage output at 765 mm Hg for the water bath temperatures noted.

TABLE V

Temperature Sensitivity of Pressure Transducers at

P = 765 mm Hg

Unit	1	3	5	7	8	9
V / ^{o}C	+0.052	-0.070	+0.009	-0.003	-0.008	-0.002

Analysis of pressure transducer calibration data was performed by the same technique used in temperature transducer analysis. The computer program and complete calibration report is available at the address given earlier. A summary of performance is given in Table VI.

TABLE VI

Summary of Calibration Results For

Respirometer Pressure Transducers

Unit No.	Sample Standard Deviation in mm Hg (755 to 765 mm Range)	Approximate 95% Confidence Level (mm Hg)
1	±0.260	±0.520
3	±0.430	±0.860
5	±0.047	±0.095
7	±0.025	±0.051
8	±0.146	±0.292
9	±0.024	±0.048

Units 5, 7 and 9 are very good, unit 8 is acceptable. The poor response of unit 3 is due to a mechanical hysteresis which was observed during the calibration but not subject to correction without disassembly. The poor response of Unit 1 could not be explained. No attempt was made to include temperature corrections during the calibration analysis. Except for Units 1 and 3, temperature induced error is significant only in the 4th decimal place. A calibration table is not available for the pressure transducers. It should be noted that an inverse calibration equation produced the best results, i.e.,

$$P = b_0 + b_1 V + ----$$

7.0 POTATO RESPIRATION DATA ANALYSIS

A computer program has been prepared for analysis of potato respiration data. It is identified as Program No. 5193, is in Fortran V language and can be obtained from the address given in section 6.5 for pressure and temperature transducer programs.

Oxygen consumption, in moles of oxygen, is calculated from Boyle's Law from the pressure and temperature data.

The output voltage at a time t (call it $v(t)$) is converted by the program into the pressure (call it $P(T)$) using the given calibration curve for the pressure transducer. This, in turn, is converted into moles using Boyle's Law for gases $P V = N R T$, so that at time t, the number of moles of gas in the chamber is:

$$N(t) = (R / V) \cdot (P(t) / T(t))$$

where V is the given volume of the chamber with all components mounted, R is the gas law constant, and T (t), is the temperature at time t in ^{o}K. The more exact Van der Walls equation of state for gases

$$(P + a N^2 / V^2)(V - b N) = N R T$$

has also been used with a difference in the value of the number of moles of gas found in the third place of the difference in the number of moles from one time to the next. If this difference is too great and/or a more accurate equation of state is suggested for the temperature region of interest, the change can be made simply. The Boyle's Law subroutine and the Van der Waal's subroutine are interchangeable.

Assuming no other gaseous change and little or no change in the volume of the chamber, the amount of oxygen consumed from a time t_{i-1} to a time t_i would be $\Delta N_i = N(t_i) - N(t_{i-1})$. this quantity is then plotted versus the time.

The quantities

$$\Delta V_i = V(t_i) - V(t_{i-1})$$

$$\Delta P_i = P(t_i) - P(t_{i-1})$$

$$\Delta T_i = T(t_i) - T(t_{i-1})$$

are also plotted versus time for comparison purposes. A lagged sample cross-covariance between the change in moles (ΔN_i) and the change in temperature (ΔT_i) can be made

$$C_j = \sum_{i=j+1}^{m} \Delta N_i \, \Delta T_{i-j}$$

where m is the number of samples. The calculation of sample spectra can also be included. Also, for comparison purposes, lagged sample cross-covariances can be calculated between ΔV_i and ΔT_i and between ΔP_i and ΔT_i.

Analysis has been performed for several experimental runs performed using bench mounted flight hardware units. Because adequate temperature control was not available at the time, the rhythms observed are almost certainly due to room temperature induced artifacts.

REFERENCES

1. "Final Technical Report, NASA Contract NASw-870" Space/Defense Corporation Report TR71-102, dated 8 January 1971.

2. Space/Defense Corporation Technical Proposal P69-136, dated 27 May 1969, "Periodicity of Potato Respiration: A Proposal for Experiments in Space Flight."

3. "Potato Respirometer Special Test Program," Space/Defense Corporation Report TR70-128, dated 1 December 1970.

4. "Design and Development of a Microbiological Respirometer with Space Flight Applications," P. C. Taudvin, B. W. Pince and J. M. Paros, presented at the 17th Annual ISA Aerospace Instrumentation Symposium, May 1971.