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Prototype Operations Manual

MELISSA



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Acronyms and Abbreviations

CSA: Canadian Space Agency
CESRF: Controlled Environment Systems Research Facility
DACS: Data Acquisition and Control System
EC: Electrical conductivity
ESA: European Space Agency
HPC: Higher Plant Chamber
HPLC: High Pressure Liquid Chromatography
HPS: High-Pressure Sodium (lamps)
IRGA: Infra-Red Gas Analyzer
MELISSA: Micro-Ecological Life Support System Alternative
MFC: Mass flow controller
MH: Metal-Halide (lamps)
MPP: MELISSA Pilot Plant
NCER: Net Carbon Exchange Rate
PPF: Photosynthetic Photon Flux
SOP: Standard Operating Procedure
TN: Technical Note
UAB: University “Autonoma” of Barcelona
UBP: University Blaise-Pascal
UoG: University of Guelph
VPD: Vapour Pressure Deficit

1. Introduction

This document outlines the operational procedures of the prototype plant growth chamber HPC1, and describes control system operation, calibration and cleaning operations.

2. Argus Control System Operation

2.1. Overview

Environment control is based on a distributed real-time stand-alone microcontroller system designed by Argus Control Systems Ltd. (White Rock, British Columbia, Appendix 1). The system consists of a dedicated network of stand-alone controllers and expansion Input/Output hardware. For the purpose of control, all sensor readings are sampled once every second while data point collection frequency is every 10 seconds, with a user selectable data presentation interval. The operator interface for the DACS is provided through a system access and management program (Argus for Windows) running under the Microsoft Windows XP operating system. All user configurable settings on the controllers are accessed using this program. The PC component of the Argus system is not used for real time control, and failure of the PC has no consequence on chamber control. The DACS makes use of numerous setpoint and feed forward algorithms to control temperature, VPD, nutrient solution composition, lighting, and carbon dioxide concentration.

2.2. Basic User Interfaces

There are four primary user screens in this implementation of the Argus Titan operator interface. The main **Home** screen (Illustration 1), the **HPC1 System Overview** screen (Illustration 2), the **HPC1 System Control** screen (Illustration 3), and the **Data acquisition and Graphing** screen (Illustration 4). The Home screen is primarily an administrative access page. Users should not alter any of the control settings that are available in this window. The HPC1 System Overview shows the current environmental parameters including chamber and lamp loft temperature, humidity, VPD, EC, pH, nutrient system status, PAR level, lamp status, CO₂ concentration, and chamber pressure. The System Control screen is used for viewing and changing the HPC1 environment control parameters.

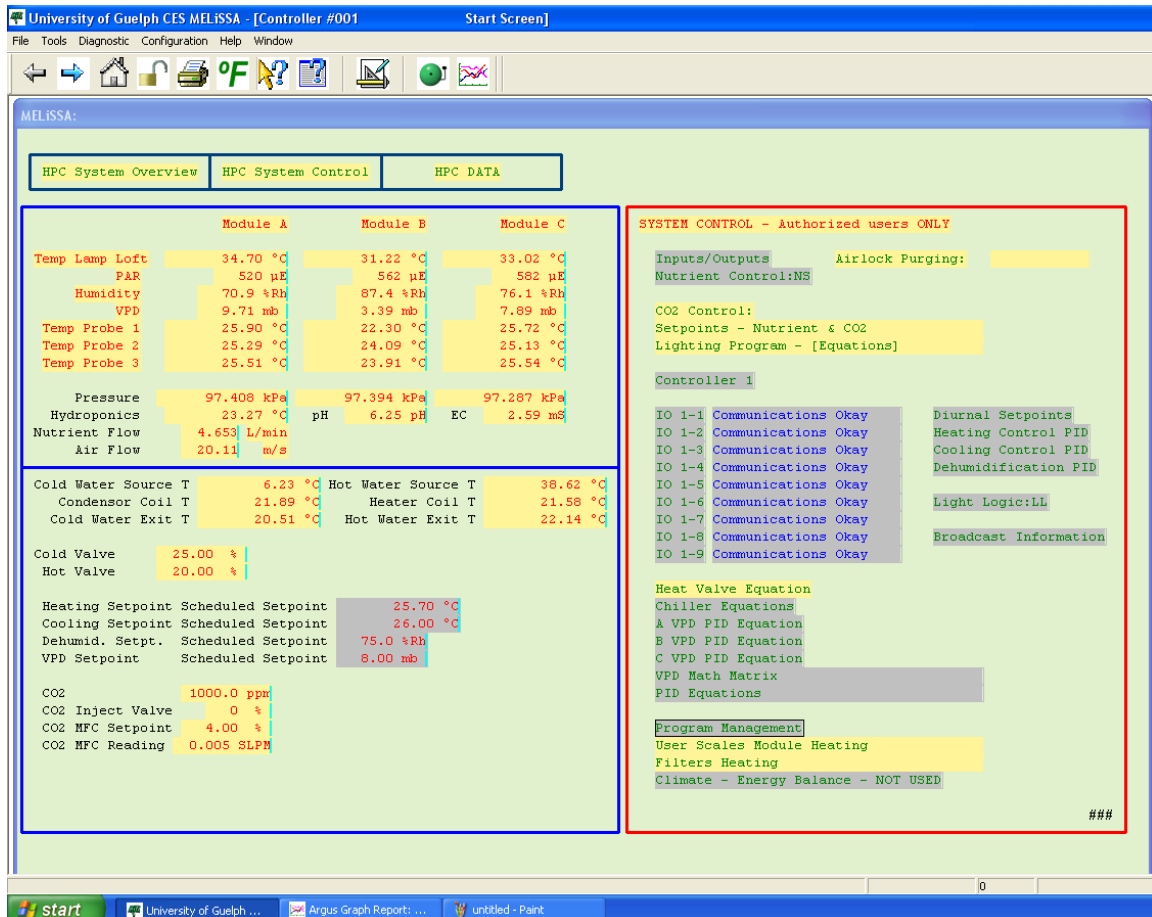


Illustration 1: Aarus Titan Home screen

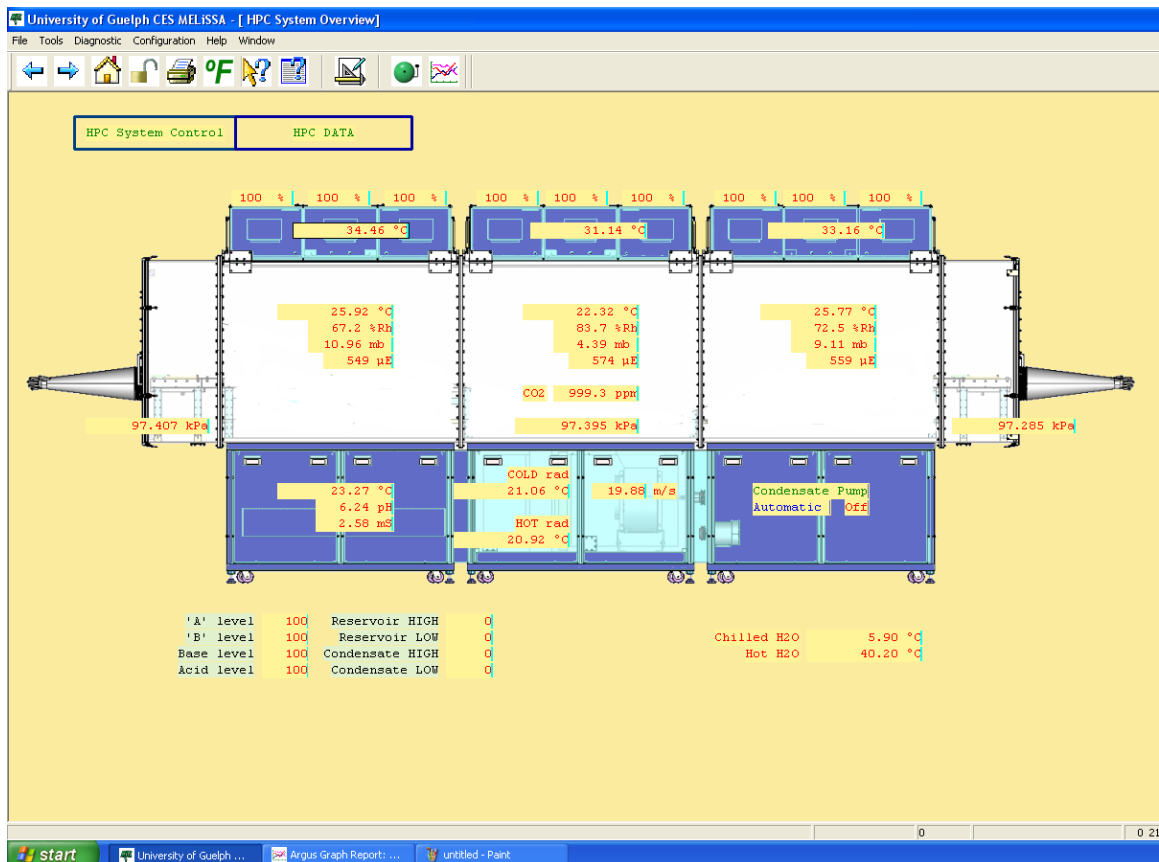


Illustration 2: HPC1 System Overview screen.

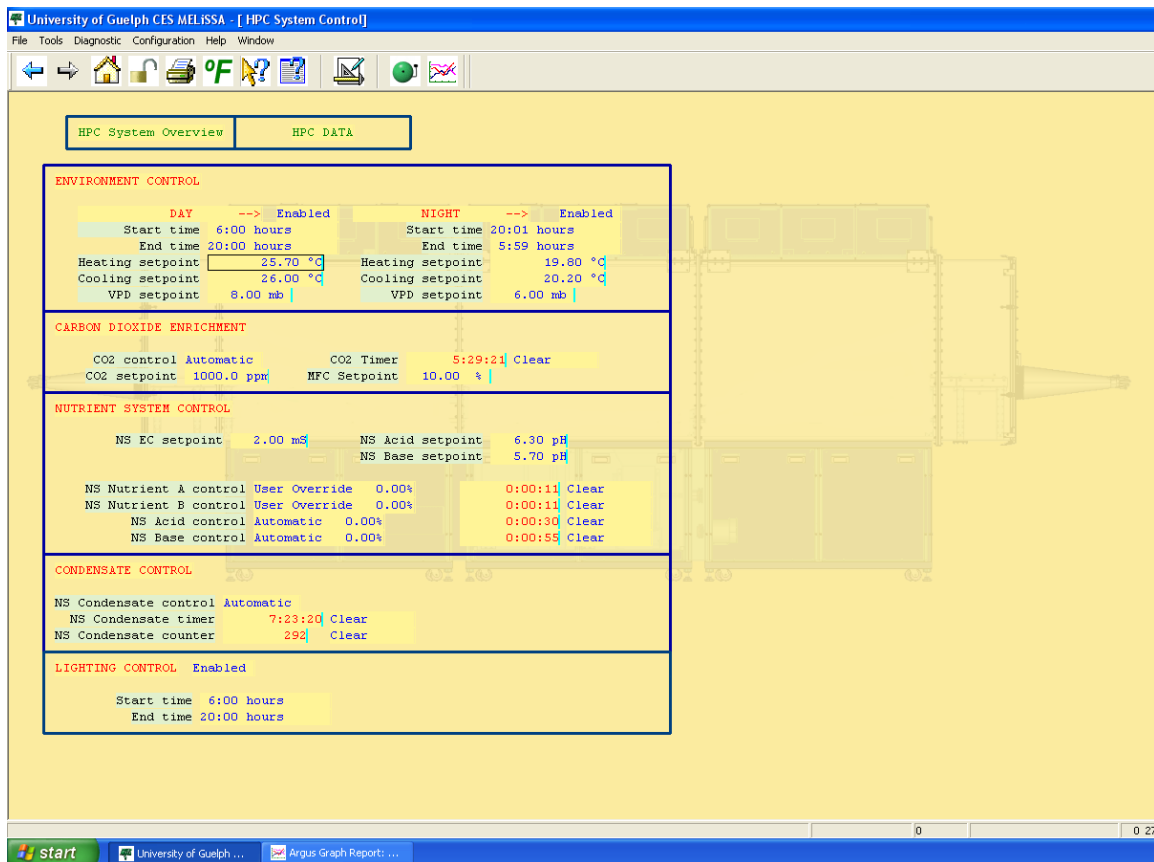


Illustration 3: HPC1 System Control screen

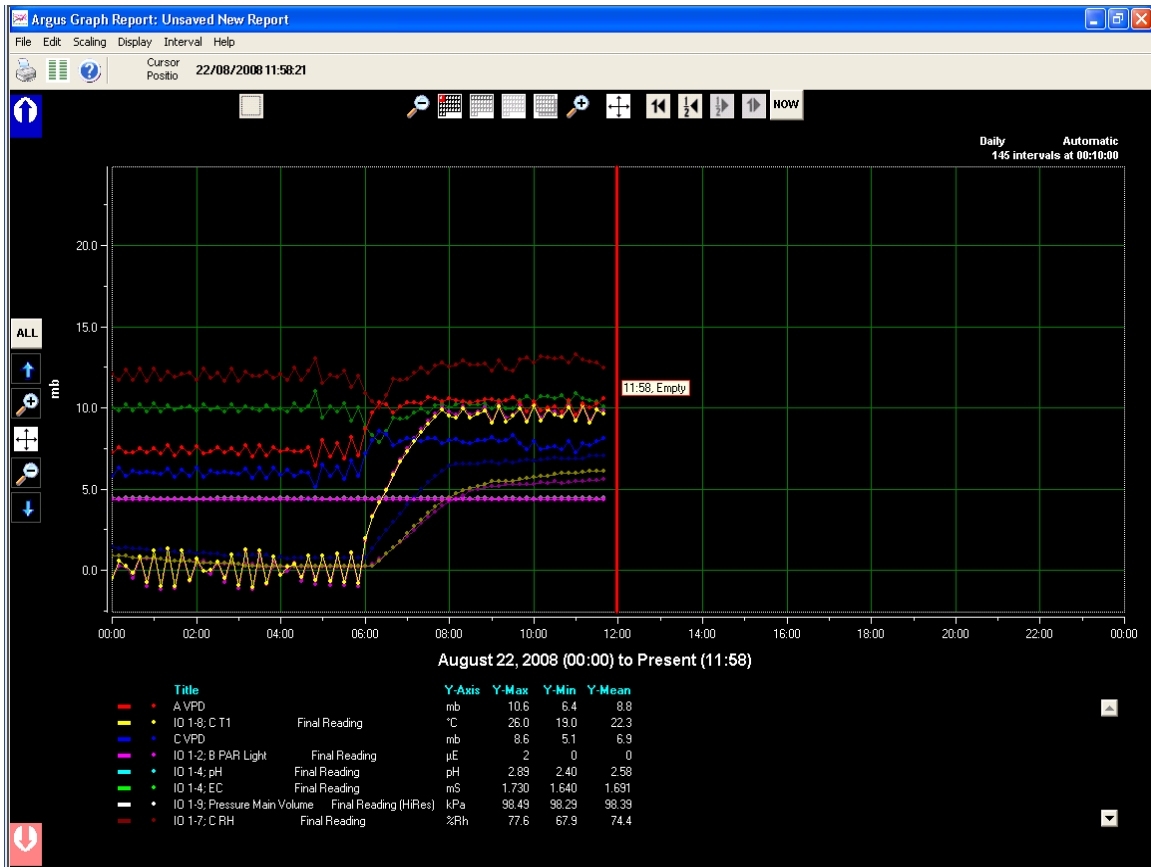


Illustration 4: Argus Data Acquisition and Graphing window

2.3. System Login

In order to change environment parameters, the HPC1 user must log into the Argus Titan system. To access the login window, click on the 'lock' icon located at the top of the main window (Illustration 4). In the login screen, the following username and password are required:

Username: UABcontrol
Password: chlorophyll

Administrative mode will be automatically unlocked after a user selected 'Timeout' period. The Timeout can be set between 3 and 240 minutes.

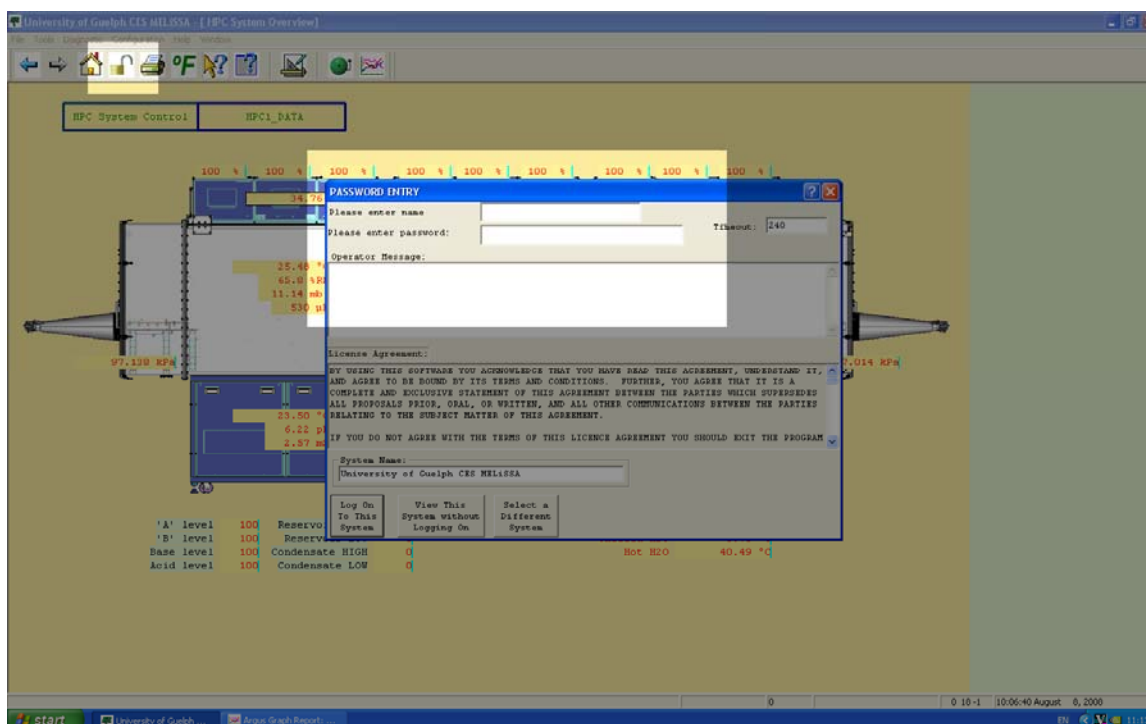


Illustration 5: Administrative login window.

2.4. Temperature and VPD Control

Control of temperature and VPD is performed by recirculating chamber air with a variable speed blower through chilled water (5 °C) and hot water (50 °C) heat exchange coils located in the central chamber unit. The cold exchange coil is used to control the VPD, while the hot exchange coil is used to reheat the cooled air to regulate the final desired temperature.

To change temperature settings, click on the parameter boxes located in the 'Environment Control' section of the HPC1 Systems Control screen (Illustration 3). Temperature control is divided into day and night periods. For isothermal operation, day control should be set to *enabled* and night control to *disabled*. For day/night temperature differences, both are set to *enable*.



For each period there is a cooling setpoint and a heating setpoint. The minimum separation between the two values should be 0.4 C. If control is set too tight, heating and cooling control will oscillate. For example, if a day time temperature of 22.0 C is desired, the heating temperature should be set to 21.8 C and the cooling temperature set to 22.2 C.

VPD is the difference (deficit) between the amount of moisture in the air and how much moisture the air can hold when saturated. VPD, rather than relative humidity, is important for proper plant growth within a controlled environment. If the VPD is too low, a film of water can form on plant leaves, making them more susceptible to rot. Conversely, if the VPD is too high, the plant needs to draw more water from its roots. The ideal range for VPD in a controlled environment is from 4.5 mb (0.45 kPa) to 12.5 mb (1.25 kPa). Generally, a setpoint of between 8 mb (0.8 kPa) and 10 mb (1.0 kPa) is employed.

Day and night VPD setpoints can be changed by clicking on the VPD values on the HPC1 Control System screen. In general, values should not be set below 6 mb or over 12 mb.

2.5. Gas Composition Control (CO₂)

Chamber gas composition is controlled by analyzer feedback to the Argus control system that operates a mass flow controller for carbon dioxide enrichment. Pure gas is supplied by an external pressurized gas cylinder. The available carbon dioxide control range is between 0 and 3000 $\mu\text{mol mol}^{-1}$. There is no method employed for removal other than photosynthetic uptake.

To enable CO₂ control, change the 'CO₂ control' parameter to 'Automatic'. Disabling, which should be done prior to any system opening or whenever the chamber doors are opened to room atmosphere, is done by switching to 'Manual Off'. To change the CO₂ control setpoint, click on the 'CO₂ setpoint' value on the HPC1 Control System screen. Valid parameters are from 0 to 2000 ppm (or higher depending upon the CO₂ analyzer in use). The flow rate of the MFC is changed by altering the 'MFC setpoint' value. Valid inputs are from 0 to 100% of full scale. This roughly equals 0 to 1000 mL/min, although the exact value of the MFC flow at any given setpoint should be verified on the MFC controller screen and recorded.

Operation and maintenance of the gas analyzer should be performed according to the manufacturers user manual (Appendix 2). The MFC operation manual is presented in Appendix 3.

2.6. Nutrient Delivery System Control

The nutrient delivery system utilizes a NFT design. Water is stored in a 200 litre tank. A stainless steel pump provides sufficient pressure/volume for in-tank circulation through a sensor loop and chamber trough delivery. Return of water from the chamber is by gravity. All external storage tanks for stock nutrients, acid, and base are maintained at chamber pressure through pressure compensation lines. An electrical conductivity sensor is used to measure nutrient concentration and a pH sensor monitors solution pH. Solution composition control uses feedback algorithms provided by the DACS in response to sensor readings and user input parameters. Concentrated acid, base, and nutrient solutions are added using gravity feed from external reservoirs.

The pH and EC delivery system is enabled by changing the 'User Override 0.00%' to 'Automatic' for each of the system components (Acid, Base, Solution A, Solution B) in the Nutrient System Control section of the Argus system control screen. The reverse of this will disable the system.

To change pH and EC setpoints, click on the EC, Acid, or Base setting boxes. EC settings generally range between 1.0 and 2.0 mS, but is dependent upon the crop and its requirements. pH has separate settings for acid and base. The acid setting should be 0.3 pH units above the desired pH value, while the base should be 0.3 pH units below the desired value. If settings are too close, system oscillation will occur resulting in excessive acid/base use and over ranging EC values. For all NDS setpoints please refer to the SOP for the crop being tested.

2.7. Lighting System Control

The HPC1 lighting system consists of nine (9) lamps wired in groups of three over each chamber subsection. In the lighting control section of the Argus system control screen (Illustration 3), specify start and end times by clicking on their respective boxes. Lighting must be set to 'enabled' for automated control to work. To disable control, set the system to 'disabled'.

2.8. Graphing and Data Acquisition

Access to the Argus Graphing and Data Acquisition screen is obtained by clicking the 'HPC1 Data' box at the top of the home, system control, or system overview screens (Illustration 4).

3. Basic Chamber Operation

3.1. Exterior Doors

To open the exterior doors, unlatch the closure mechanisms in the sequence (Illustration 6). Carefully open the door using the handle provided. To close the door, push the door towards the chamber and latch the closures in the reverse order shown in the illustration.



CAUTION! The Viton™ door seal is susceptible to mechanical damage. Use care when opening and closing the external door.

ENSURE that the gloves **DO NOT** hang below the bottom door seal. Closing the door seals on a glove could damage seal and glove integrity.



Illustration 6: HPC1 exterior door latch locations. Red arrows indicate latch mechanism locations. Numbers represent the unlatching sequence.

3.2. Interior Doors

CAUTION: The magnetic seals are susceptible to mechanical damage. Use care when opening and closing the inner door. Keep objects that may fall on the seals away from the door opening..

If the exterior doors must remain closed during this operation, use the glove boxes to reach inside and follow these procedures:

To open:

1. Pull the bottom magnetic seal downwards until it is free of the door material (Illustration 7).
2. Pull the right magnetic seal towards the right side of the chamber until it is free of the door material.
3. Pull the left magnetic seal towards the right side of the chamber until it is free of the door material.
4. Using the door manipulation bar provided, hook the top stainless steel door insert into the top hooks that are furthest away from you.
5. Hook the lower stainless steel door insert into the top hooks closest to you.

To close:

1. Be sure the door opening is free of obstructions.
2. Carefully lower the front and back stainless steel door inserts so that the door hangs freely in the opening.
3. Hold the left bottom corner tight by pulling down and to the left.
4. Close the left magnetic seal.
5. Hold the right bottom corner tight by pulling down and to the right.
6. Close the left magnetic seal.
7. Close the centre magnetic seal.

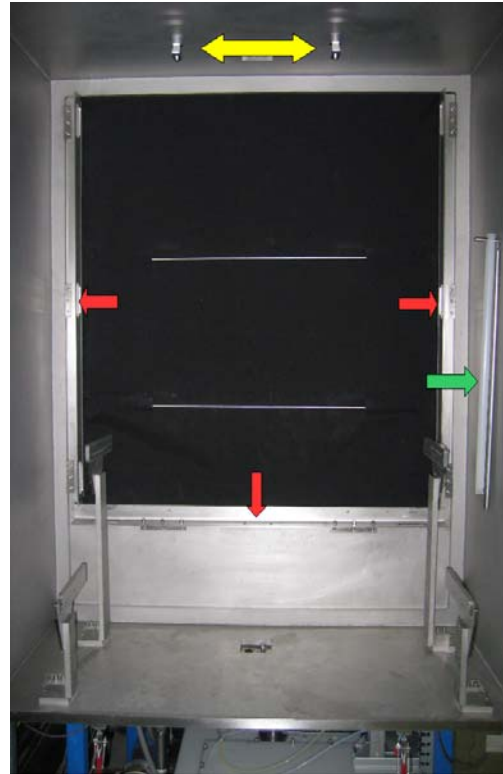


Illustration 7: HPC1 air lock interior door. Red arrows indicate the magnetic clamps. The green arrow shows the location of the door manipulation bar, and the yellow arrows indicate the top door hooks.

3.3. Stock Reservoir filling

To fill or refill the acid/base/nutrient stock tanks, the following procedure should be employed:

1. Prepare 3.0 litres of stock solution(s) according to the SOP for the crop.
2. Slowly tilt open the front panel housing the stock reservoirs (Illustration 8).
3. Firmly hold the sides of the reservoir being filled and unscrew the cap. **NOTE:** never open more than one reservoir at a time.
4. Carefully pour the new solution into the reservoir. **DO NOT** overfill the reservoir. **NEVER** fill beyond $\frac{3}{4}$ of the total reservoir capacity.
5. Firmly hold the sides of the filled reservoir and firmly tighten the cap.
6. Repeat the procedure, if required, with the remaining stock solutions.
7. Slowly close the front panel.



CAUTION! Be advised that acids and bases can cause serious injury. Always use caution and wear proper protective equipment when working with either substance. See your laboratory safety procedures for more information on working with acids and bases.

NEVER work with both acid and base stock tanks open at the same time. Fill one, close it, then fill the other. **DO NOT MIX THEM UP!!**



Illustration 8: Location of the stock solution reservoirs. Note that the front panel tilts forward to allow access.



Illustration 11: Stock solution isolation valves are located on top of the NDS main reservoir.



Illustration 10: NDS pump controller.



Illustration 9: Blower remote interface.

3.4. Stock Reservoir Isolation

To manually isolate the stock reservoirs from the rest of the NDS, close the single air compensation valve and the four manual acid/base/nutrient valves located on top of the NDS main tank (Illustration 9).

3.5. NDS Pump Operation

The NDS pump controller is located on the left side of the NDS compartment of HPC1.

3.6. Blower Operation

The HPC1 blower is manually operated using a remote power interface. To turn the fan on, move the power interface switch to the 'ON' position and adjust the blower speed to a reading of '41'. Turning it off is the reverse of this procedure. The fan must remain on during day and night periods to ensure proper heating and cooling of the chamber.

4. Crop Culture Procedures

Crop culture procedures are specific to the species/cultivar selection, length of crop cycle, and number of stages in continuous production. Please refer to the SOP or TN for different cropping procedures. Harvest and analytical procedures will be included in the cultivation SOPs.

Required information contained within an SOP include:

- Crop variety and planting density

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- Day/Night period length
- Temperature setpoint(s)
- VPD setpoint(s)
- CO₂ concentration setpoint
- Nutrient main tank and A/B recipes – EC setpoint
- Acid and Base type and concentration – pH setpoint
- Crop growing period
- Planting media and planting details
- Harvest interval and harvest details
- Analytical procedures and requirements

5.Planting/Harvesting Procedures

The following are the general procedures for adding/removing growing trays to/from HPC1. Only one tray is added or removed at a time. There are three procedural options: planting with no harvest, which can occur at the beginning of an experiment when the chamber is not yet at capacity; harvesting with no planting, which can occur when the experiment is nearing its conclusion and no further plants are being introduced to the chamber; and a complete plant/harvest cycle. Prior to any planting procedure, the chamber must be equipped with a full compliment of troughs.

These steps should be performed as quickly as possible if it takes place during the chamber day period. If it expected to take longer than 15 minutes, turn off the lamps so the plants do not dry out. Because of this, dark period harvest/planting is preferred. HPC1 day/night periods can be offset from earth day/night periods to account for staffing availability during normal working hours.

5.1. Plant (no harvest)

1. Prepare the growing tray as per crop SOP.
2. Close the chamber trough feed valve.
3. Ensure interior air lock door seals at the planting and harvesting ends of the chamber are closed.
4. Open the exterior planting air lock door.
5. Place the single growing trough with seedlings into the air lock on the upper trough support. Ensure the trough drain hole is on the same side as the interior drainage trough.
6. Close the exterior planting air lock door and ensure a proper seal.
7. Purge the air lock volume with the calibrated air stream (Planting purge sequence).
8. Using the glove box, open the interior planting and harvest air lock doors.
9. Operators must now be manning the glove boxes at both ends of HPC1.
10. On the planting end, fasten to the troughs already on the conveyer and push the trough forward towards the harvest end.
11. On the harvest end, unhook the empty trough and place it on the lower trough support.
12. Ensure the NDS spouts are roughly centred on the troughs within the chamber.
13. Close the interior air lock doors on the planting and harvesting ends.
14. Open the harvest end exterior door and remove the empty trough.
15. Close the exterior harvest air lock door and ensure a proper seal.
16. Purge the harvest air lock (Harvest purge sequence).
17. Open the chamber trough feed valve to reactivate the irrigation system.

5.2. Harvest (no planting)

1. Close the chamber trough feed valve.
2. Ensure interior air lock door seals at the planting and harvesting ends of the chamber are closed.
3. Open the exterior planting air lock door.
4. Place a single empty growing trough into the air lock on the upper trough support. Ensure the trough drain hole is on the same side as the interior drainage trough.
5. Close the exterior planting air lock door and ensure a proper seal.
6. Purge the air lock volume with the calibrated air stream (Planting purge sequence).
7. Using the glove boxes, open the interior planting and harvest air lock doors.
8. Operators must now be manning the glove boxes at both ends of HPC1.
9. On the planting end, fasten to the troughs already on the conveyer and push the trough forward towards the harvest end.
10. On the harvest end, unhook the trough to be harvested and place it on the lower support.
11. Ensure the NDS spouts are roughly centred on the troughs within the chamber.
12. Close the interior air lock doors on the planting and harvesting ends.
13. Open the harvest end exterior door and remove the trough. Harvest the crop as per the SOP.
14. Purge the harvest air lock (Harvest purge sequence).
15. Close the exterior harvest air lock door and ensure a proper seal.
16. Open the chamber trough feed valve to reactivate the irrigation system.

5.3. Plant/Harvest

1. Prepare the growing tray as per crop SOP.
2. Close the chamber trough feed valve.
3. Ensure interior air lock door seals at the planting and harvesting ends of the chamber are closed.
4. Open the exterior planting air lock door.
5. Place the single growing trough with seedlings into the air lock on the upper trough support. Ensure the trough drain hole is on the same side as the interior drainage trough.
6. Close the exterior planting air lock door and ensure a proper seal.
7. Purge the air lock volume with the calibrated air stream (Planting purge sequence).
8. Using the glove box, open the interior planting and harvest air lock doors.
9. Operators must now be manning the glove boxes at both ends of HPC1.
10. On the planting end, fasten to the troughs already on the conveyer and push the trough forward towards the harvest end.
11. On the harvest end, unhook the full trough and place it on the lower trough support.
12. Ensure the NDS spouts are roughly centred on the troughs within the chamber.
13. Close the interior air lock doors on the planting and harvesting ends.
14. Open the harvest end exterior door and remove the trough. Harvest the crop as per the SOP.
15. Close the exterior harvest air lock door and ensure a proper seal.
16. Purge the harvest air lock (Harvest purge sequence).
17. Open the chamber trough feed valve to reactivate the irrigation system.

6. Daily Operations Checklist

- ✓ Check temperature, VPD, CO₂, pH, EC status over the last 24 hours in graphing program. Ensure that there have been no deviations since the last check.

- ✓ Check status of bottled CO₂. Replace tank when pressure falls below 500 psi.
- ✓ Check Acid/Base/Nutrient A/B level status.
- ✓ Check condensate level status.
- ✓ Check NDS tank level status.
- ✓ Check nutrient flow status.
- ✓ Check air flow and blower operation.
- ✓ Visually inspect plant growth and development.
- ✓ Check that chilled and hot water temperatures are within operating levels.

7. System Calibration

Most of the HPC1 subsystems require periodic calibration to maintain proper functioning.

7.1. pH and Electrical Conductivity (EC)

1. In the Argus Control System, change the acid, base, and nutrient A/B control settings from 'Automatic' to 'User Override 0.00%'. This is to ensure that the system does not activate during the calibration procedure.
2. Close the two isolation valves on the nutrient system bypass loop (Illustration 5).
3. Pour known standards of pH and EC into 250 mL beakers. Use two point calibrations with solutions of 4.00 and 10.00 pH units, and ~500 and ~3000 uS.
4. Carefully unscrew (counterclockwise to loosen) and remove the EC and pH sensors.
5. Place the probes into their respective solutions and follow the calibration procedures outlined in the operation manuals (Appendix 4)
6. Rinse the probes, apply Teflon tape to the threads, and screw them back into the nutrient system bypass manifold.
7. Reopen the bypass isolation valves to the same position as they were before calibration.
8. Return the Argus Control System settings to 'Automatic'.

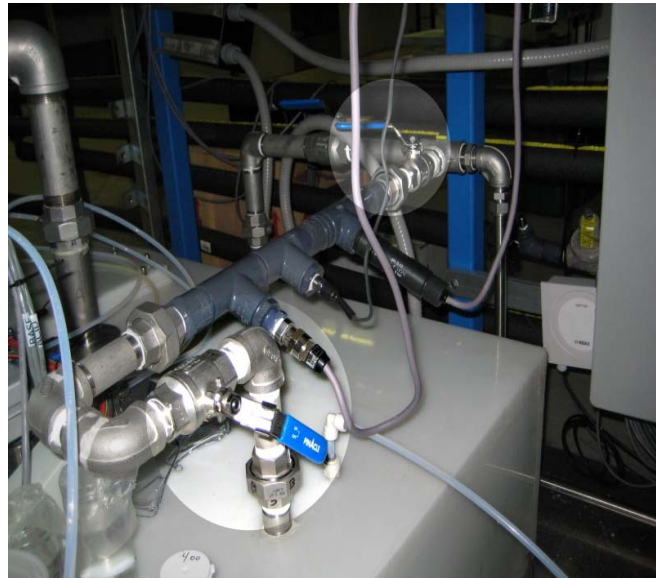


Illustration 12: Location of shut off valves for pH/EC calibration procedure.

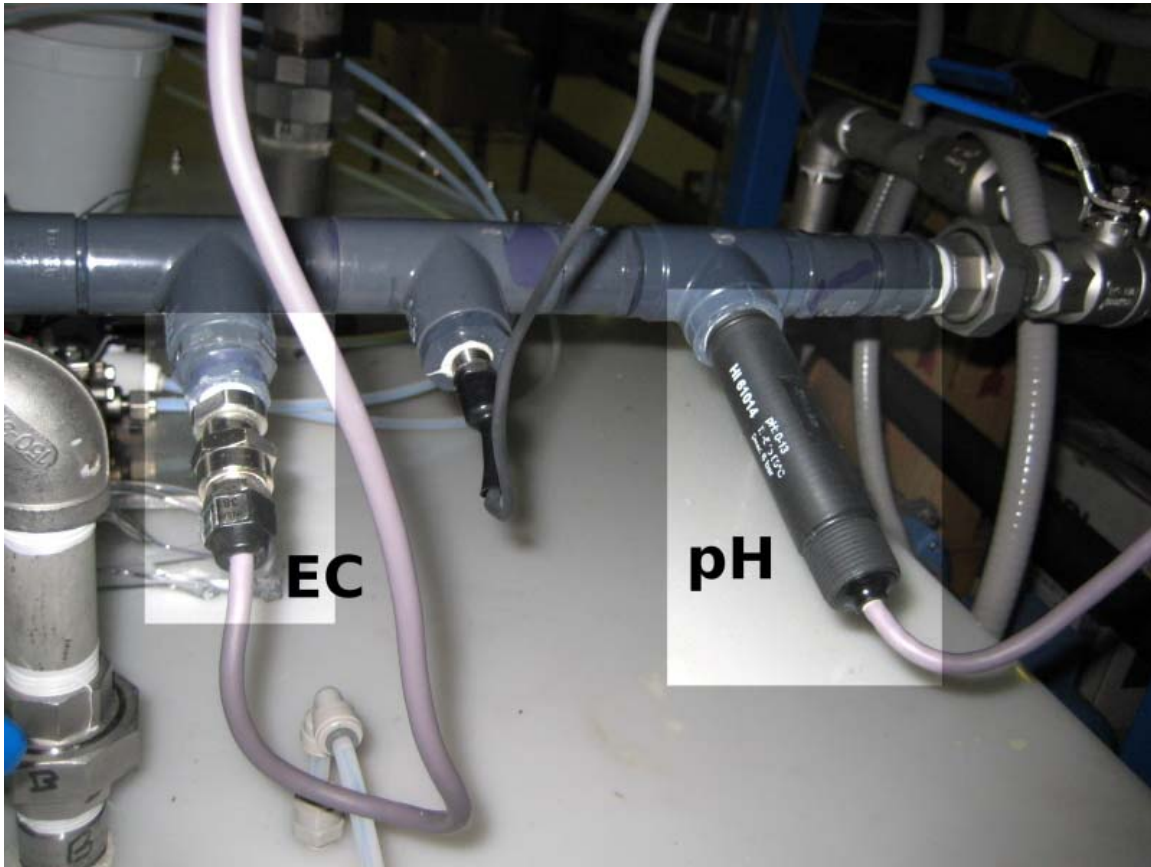


Illustration 13: Location of EC/pH sensors.

7.2. Nutrient Delivery System Calibration

NDS system calibration should be performed at the beginning and end of each experiment.

1. Empty the main NDS tank of all fluid using the drain valve. Leave valve open once drained.
2. Empty and dispose of acid, base, and nutrient stock solutions according to laboratory protocol for hazardous waste materials.
3. At the Argus relay panel, switch the acid, base, nutrient A and nutrient B relays to 'ON'.
4. Fill each reservoir with 3 litres of clean water and allow to drain into the main tank. This is to remove any residual acid/base/nutrient solution.
5. Repeat rinsing two more times with 3 litres of clean water.
6. Open the top of the NDS reservoir. Ensure that the water has been completely removed from the tank. Use a wet vacuum if required to accelerate the process.
7. Set the acid/base/nutrient relays to the 'OFF' position.
8. Place a clean and dry 2L beaker beneath the acid/base/nutrient outlets within the NDS reservoir.
9. Fill the acid/base/nutrient stock tanks with 3 litres of clean water.
10. Using a timer, switch the acid relay from 'OFF' to 'ON' for 30 seconds.

11. Collect, measure, and record the amount of water released into the beaker using a 100 mL volumetric flask.
12. Repeat 10 until no more water flows from the stock tank into the 2L beaker.
13. Repeat this procedure for each stock tank.
14. Use the collected data to estimate the flow rate for each container.

7.3. Condensate System Calibration

1. At the Argus relay panel, switch the condensate pump relay to 'OFF'.
2. Fill the condensate reservoir with clean water to approximately half full.
3. Disconnect the quick connect pump line from the main NDS reservoir.
4. Place the pump line outlet into a 500 mL volumetric flask.
5. Turn the condensate pump relay to the 'ON' position and time, using a stopwatch, the number of seconds required to fill the volumetric flask to 500 mL.
6. Use the collected data to estimate the pump flow rate.

7.4. CO₂/O₂



CAUTION! Carbon dioxide is a colourless, odourless, faintly acidic-tasting, and non-flammable gas at room temperature.


Releasing carbon dioxide gas in a confined or unventilated area can lower the concentration of oxygen to a level that is immediately dangerous to life or health.

Follow laboratory safety protocols when working with bottled gases.

Operation and maintenance of the gas analyzer should be performed according to the manufacturers user manual (Appendix 2). The MFC operation manual is presented in Appendix 3.

8. Basic Maintenance Tasks

8.1. Bulb replacement

	<p>CAUTION! Lamps can be extremely HOT if they have been recently on. Before attempting replacement, wait at least one hour for lamps to cool sufficiently.</p>
	<p>CAUTION! Lamp replacement requires the use of ladders and working at an elevated level. Use proper safety procedures. Never work alone when changing lamps.</p>

When required, bulbs should be replaced by following this procedure:

1. Turn off lighting system main circuit breaker and lock out according to laboratory protocol.
2. Using an approved mobile warehouse step, climb to a point where your knees are level with the lamp loft handle (Illustration 14).
3. Lift the lamp loft using the handle and carefully raise and push it back until it is securely in place.
4. Unscrew the lamp(s) and replace with a new lamp. Use gloves to avoid transferring oils to the lamp surface (Illustration 15).
5. Carefully pull back on the lamp loft handle and gently lower it back into place.
6. Remove the mobile warehouse step.
7. Unlock and turn on the lamp lighting main electric circuit.



Illustration 15: Lamp locations within the lamp loft.



Illustration 14: Location of the lamp loft and lifting handle (yellow arrow).

8.2. Cleaning



CAUTION! Cleaning chemicals can be extremely dangerous. Be sure to read and understand your MSDS and follow all laboratory safety protocols when handling chemicals.

CAUTION! Bleach, alcohol, and ozone can be dangerous in confined spaces. Be sure the air handling system is operational and all exterior and interior air lock doors are open when working within the chamber. NEVER work inside HPC1 without adequate supervision.

To ensure a minimal level of contamination with algae, biofilms, and other microorganisms, thorough cleaning should be performed at the end of each experiment.

Exterior

The exterior surfaces can be cleaned with warm water and mild dish washing soap.

NDS Plumbing system

1. Empty the main NDS reservoir and fill with 180 litres of a 10 ppm ozone solution. (Note: a 0.5% bleach solution can be substituted for ozone, however ozone is the preferred method).
2. Run the NDS system with plant troughs in all positions for a period of up to 24 hours.
3. Drain the NDS reservoir and fill with clean water.
4. Run the NDS system for up to 24 hours.
5. Drain the NDS system and fill with clean water.
6. Run the NDS system for 6 hours.
7. Drain the NDS system and fill with clean water.
8. Run the NDS system for 1 hour.
9. Drain the NDS system.
10. Open the NDS reservoir cover and wipe down the inside surfaces with clean a clean cloth.

Interior

The interior stainless steel surfaces should first be cleaned with a mild dish washing soap in warm water, followed by disinfection with a high-concentration alcohol mixture (i.e. 80% ethanol + 5% isopropanol).

Growing Troughs

Growing troughs should be removed from the chamber and cleaned with warm water containing mild dish washing soap. Surface disinfection to remove algae contamination can be done with a 5 ppm ozone solution (85% alcohol or 0.5% bleach in water can be substituted). Ideally and when possible, troughs should be soaked for a 12 hour period in a 5 ppm ozone solution. Troughs **MUST** be adequately rinsed prior to return to service.

Lamp Loft Glass Panels

MELISSA



TECHNICAL NOTE

The glass panels in the lamp loft should be cleaned with warm water and mild dish washing soap. Rinse with clean water and dry with a clean cloth.

9. Appendix 1 - Argus Titan System Description

Electronic File: Titan_System_Description.pdf

10. Appendix 2 - Operation Manual - CO₂/O₂ analyzer

The User Manual will not be available until the analyzer is delivered to UAB in Fall/Winter 2008.

11. Appendix 3 - Operation Manual - Mass Flow Controller

Electronic File: MFC.pdf

12. Appendix 4 - User Manuals - pH and EC probes

Electronic File: manHI8614_ph_transmitter_manual.pdf
manHI8931-2_EC_transmitter_manual.pdf