Biological Imaging Capability in the ABRS Facility on ISS

David R. Cox

ASGSB 2010

11/7/10

Agenda

- ABRS Background
- APEX-Cambium Background
- ABRS Green Florescent Protein Imaging System
- GIS Development (for TAGES)
- GIS Demonstrated Capability (w/ TAGES)
- GIS Availability
- GIS Enhancements?

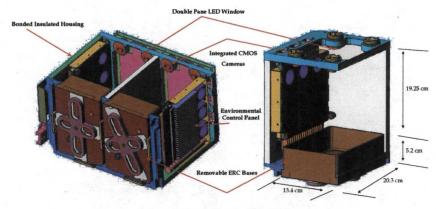
ABRS Background

- The Advanced Biological Research System (ABRS) was developed as an environmental control chamber for ISS as an EXPRESS Sub-Rack Facility
- ABRS has two independently controlled Experiment Research Chambers (ERCs) with temperature, relative humidity, carbon dioxide

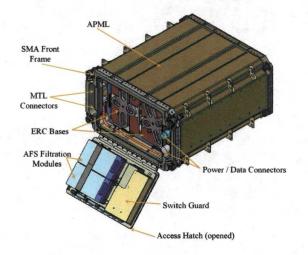
Biological Flight Systems Hardware Development Key Projects

ABRS Highlights

- Single Middeck Locker Equivalent (MLE)
- Capable of launching and returning live plants
- STS Middeck and ISS EXPRESS Rack compatible
- Designed for extended duration on ISS with ORU refurbishment and external water addition
- First flight will include GFP imaging capability
- Provides programmable LED lighting, Temperature, Relative Humidity and Carbon Dioxide control, ethylene scrubbing and water recycling

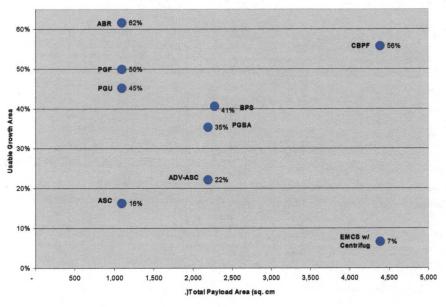


Environmental Research Chamber (ERC) - Details



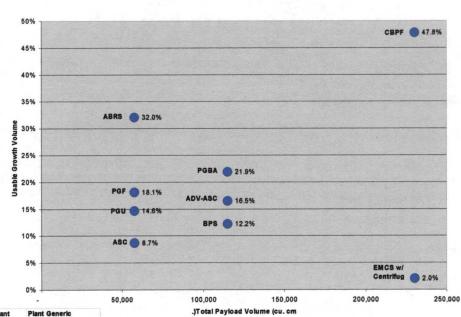
Plant Flight H/W Comparison





ABRS Growth area and volumetric efficiencies due to miniaturization and innovative technology improvements

Usable Volume Comparison



System Performance Specifications	Advanced Biological Research System (ABRS)	European Modular Cultivation System (EMCS)	Biomass Production System (BPS)	Advanced Astroculture (ADVASC)	Commercial Plant Biotechnology Facility (CPBF)	Plant Generic Bioprocessing Apparatus (PGBA)
Outer Envelope	1 middeck locker	quad locker	double middeck locker	double middeck locker	quad locker	double middeck locker
Growing Area	16.0 (W) x 21.1 (D) cm	6.0 (W) x 6.0 (D) cm	15.2 (W) x 15.2 (D) cm	unknown	unknown	25.4 (W) x 30.5 (D) cn
Growing Area Height	27.0 cm	16.0 cm	15.2 cm	34.5 cm	45.0 cm	32.5 cm
Total Growing Volume	675 cm3	288 cm3	924 cm3	486 cm3	2443 cm3	775 cm3
No. of Growth Chambers	2	8 (4 per centrifuge rotor)	4	1	1	1

ABRS is a third generation plant growth system based on PGU and PGF knowledge

ABRS uses half (or less) of the mass, volume and power of other plant growth systems

Advanced Biological Research System (ABRS) Sustaining Engineering / Maintenance WBS: TBD



PI: N/A PS: N/A

PM: David R. Cox

Engineering Team: Life Sciences Services Contract (LSSC) at KSC

Objective:

- Provide on-going maintenance and sustaining engineering for ABRS flight and ground units
- Ensure enhancements made to on-orbit unit(s) are successful and are mimicked in the ground unit(s)
- Provide Certificate of Flight Readiness (CoFR) and Flight Safety documentation for all ISS Stage reviews

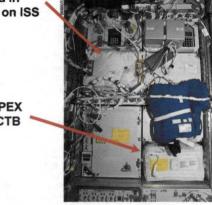
Relevance/Impact:

- Maintain ABRS at full capability to enable continued use of microgravity and other characteristics of space to enhance understanding of fundamental biological processes
- Retain ABRS demonstrated scientific return through return of biological specimens as well as via telescience using the Green Fluorescent Protein (GFP) Imaging System (GIS)
- Ensure research integrity is preserved through proper use of ground controls

Development Approach:

- Use generally accepted engineering practices and research standards to maintain ABRS flight and ground hardware and software
- Follow ISS Payloads Office (OZ) templates to meet required CoFR and Flight Safety deliverables

ABRS Installed in EXPRESS Rack 2 on ISS



ABRS and APEX Stowage in CTB

ISS Resource Requirements

Accommodation (carrier)	EXPRESS Rack 2 on ISS		
Upmass (kg) (w/o packing factor)	N/A		
Volume (m³) (w/o packing factor)	ABRS Facility 1 MLE & ABRS / APEX Stowage 1 CTB		
Power (kw) (peak)	0.320		
Crew Time (hrs) (installation/operations)	TBD		
Launch/Increment	ULF3/Increment 21 and subs		

Project Life Cycle Schedule

Milestones	CDR	Safety	FHA	Launch	Ops	Return	Final Report
Actual	Jan 2005	Mar 2009	Nov 2009	STS-129/ULF3	TBD	N/A	N/A

Advanced Plant Experiments on ISS – Cambium (APEX-Cambium)

WBS: 825080.04.05.20.05

Pls: CSA: Rodney Savidge, Ph.D., Univ. of New Brunswick, Jean Beaulieu, Ph.D., Natural Resources of Canada NASA: Robert Ferl & Anna-Lisa Paul, Ph.Ds., Univ. of Florida

PS: Howard G. Levine, Ph.D., NASA-KSC

PM: David R. Cox, NASA-KSC

Engineering Team: Life Sciences Services Contract (LSSC) at KSC

Objectives:

- Launch and execute the APEX-Cambium payload operations in conjunction with the Canadian Space Agency (CSA) and International Space Life Sciences Working Group (ISLSWG) agreements.
- Cambium: Determine the role of gravity in Cambium wood cell development.
- Transgenic Arabidopsis Gene Expression System (TAGES): Demonstrate nondestructive reporter gene technology & investigate spaceflight plant stress.

Relevance/Impact:

- APEX-Cambium payload will provide NASA and the ISS community a permanent controlled environment capability (the Advanced Biological Research System (ABRS)) to support growth of various organisms (i.e. whole plants). Also, as a result of the TAGES experiment Green Florescent Protein (GFP) imager development, ISS partners will benefit from a modern biological analysis capability that can provide real time non-destructive gene expression data which can ultimately optimize ISS microgravity biological experimentation and greatly reduce required specimen downmass.
- The Cambium experiment will provide the pulp & paper and construction industries insight into the fundamental mechanisms of wood cell formation.
- The TAGES experiment will demonstrate non-destructive GFP reporter gene technology and improve the understanding of spaceflight stresses on terrestrial organisms.

Development Approach:

- APEX-Cambium completed the Advanced Biological Research System (ABRS) final ISS verifications, assembly, Launch and initial on-orbit operations.
- Design, develop and test a Green Florescent Protein (GFP) imaging system for use in ABRS.















Clockwise from Top Left: Advanced Biological Research Sys. (ABRS), Dr. Savidge with Willow trees, ABRS Green Florescent Protein (GFP) Imaging System, TAGES plant GFP image, Dr. Beaulieu with Spruce trees, ABRS in EXPRESS Rack 2, Drs. Ferl & Paul with TAGES plants.

ISS Resource Requirements

Accommodation (carrier)	Middeck locker and CTB		
Upmass (kg) (w/o packing factor)	38.9		
Volume (m³) (w/o packing factor)	0.073		
Power (kw) (peak)	0.320		
Crew Time (hrs) (installation/operations)	27.0		
Launch/Increment	ULF3/Increments 21/22 & 23		

Project Life Cycle Schedule

Milestones	CDR	Ph3 FSR	PVT	Launch	Ops	Return	Final Report
Actual/ Baseline	1/2005	3/2/09	12/3/08	ULF3/STS-129	Increments 21/22 & 23	20A, 19A & ULF4	Return + 12m

Revision Date: 05/03/10

Advanced Plant Experiments on ISS – Series (APEX-02+)



Pls: TBD from targeted NRA or ILSRA PS: Howard G. Levine, Ph.D., NASA-KSC

PM: David R. Cox, NASA-KSC

Engineering Team: Life Sciences Services Contract (LSSC) at KSC

Objectives:

- Perform plant research on ISS in support of the Fundamental Space Biology (FSB) Strategic Plan.
- Leverage International Space Life Sciences Working Group (ISLSWG) and National Lab partner ISS resources to optimize ISS utilization efficiencies and maximize scientific return to the research communities.
- When appropriate, utilize the NASA Advanced Biological Research System (ABRS) EXPRESS Sub-rack Facility on ISS including its Green Florescent Protein (GFP) Imaging System.

Relevance/Impact:

- The APEX payload series is designed to study the fundamental cellular, molecular and whole plant sensing mechanisms and reactions to changes in gravity.
- In addition, the APEX payload series can help determine the viability of multi-generation developmental responses to micro and fractional gravity.
- The knowledge gained in plant systems will have direct application to closing the Water, Air and Carbon loops in long duration human space travel.

Development Approach:

- Solicit peer-reviewed research in support of the FSB Strategic Plan and Decadal Studies through NASA Research Announcements (NRAs) and International Life Sciences Research Announcements (ILSRAs).
- Partner with International Space Life Sciences Working Group (ISLSWG) and National Lab partners.
- Identify opportunities for Translational Teaming wherever possible.











Clockwise from Top Left: Canadian Astronaut Bob Thirsk harvesting Willow trees, Spruce trees on ISS, Transgenic Arabidopsis Gene Expression System (TAGES) Green Florescent Protein (GFP) image, Advanced Biological Research System (ABRS) GFP Imaging System, Astronaut Jeff Williams preserving Arabidopsis in a Kennedy Fixation Tube (KFT).

ICC Deserves Deservises and

Accommodation (carrier)	Middeck locker and CTB
Upmass (kg) (w/o packing factor)	TBD
Volume (m³) (w/o packing factor)	TBD
Power (kw) (peak)	TBD
Crew Time (hrs) (installation/operations)	TBD
Launch/Increment	TBD

Project Life Cycle Schedule

		rioj	ect Lije Cyt	LIE SCHEUUIE			
Milestones	CDR	Ph3 FSR	PVT	Launch	Ops	Return	Final Report
Actual/ Baseline	TBD	TBD	TBD	TBD	TBD	TBD	Return + 12m

Revision Date: 05/03/10

TAGES - Transgenic Arabidopsis Gene Expression System

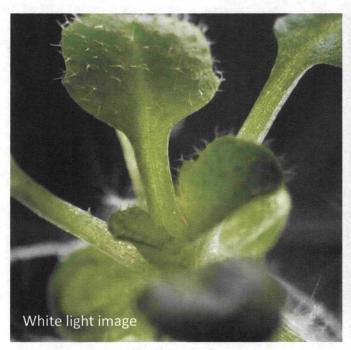
Principal Investigators:

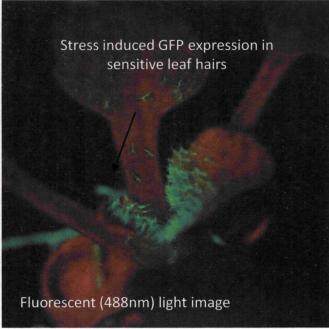
Dr. Robert J. Ferl, Professor

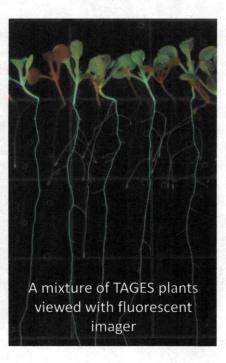
Dr. Anna-Lisa Paul, Research Associate Professor



- Using biosensor technology and telemetric imaging to capture molecular responses to spaceflight (The ABRS GIS hardware).
- Using state of the art molecular and genomic tools to evaluate changes in gene expression in returned samples.

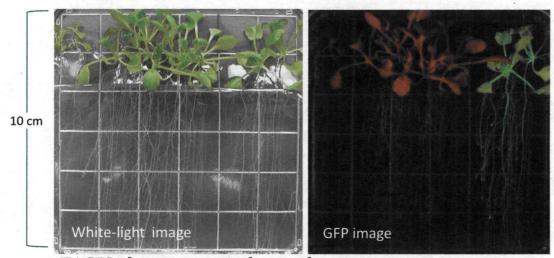






TAGES Technology is Scalable for Telemetric Data Collection in a Variety Exploration Platforms

- GFP (Green Fluorescent Protein) reporters make a fluorescent gene product gene responses captured in real-time with digital cameras.
- Digital images can be transmitted from the site of the experiment to the site of the researcher.
- Totally autonomous data collection, minimal to no crew needed:
 - Orbital (ISS) / Planetary landers / Small satellite



TAGES plants on vertical agar plates. Plants engineered to respond to reduced oxygen levels.

Peer-Reviewed Research. University based and funded by NASA: Current Funding Sources - NASA Flight (NNX07AH27G); NASA Fundamental Space Biology (NNX09AL96G); NASA MMAMA (NNX09A078G)

Orbit - Shuttle to ISS Middeck locker **Express Rack ABRS Arabidopsis** R.J. Ferl / A-L. Paul

Background

- APEX-Cambium payload includes the NASA sponsored Transgenic Arabidopsis Gene Expression System (TAGES) experiment.
- TAGES plants are genetically modified
 Arabidopsis utilizing a Green Florescent Protein
 (GFP) [naturally found in jellyfish] as a non-destructive plant stress reporting mechanism.
- Based on the TAGES requirements, a piece of Experiment Unique Equipment (EUE) was designed to fit inside one ABRS Experiment Research Chamber (ERC) to capture, store and transmit GFP images of the TAGES plants to the ground in near real time (telemetric science!)
- GIS worked flawlessly on ISS Stage ULF3.





Advanced Plant Experiments on orbit-Cambium















Natural Resources

Ressources naturelles



CSA: ILSRA-04-122

NASA: 98-HEDS-02-299

SCIENCE TEAM

Rodney Savidge, Ph.D. Univ. of New Brunswick Robert Ferl, Ph.D.

Jean Beaulieu, Ph.D. Anna-Lisa Paul, Ph.D. Natural Resources Canada

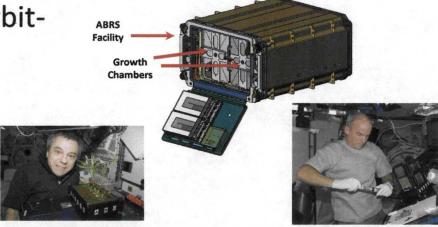
Univ. of Florida

RESEARCH OBJECTIVES

There are two plant experiments in the APEX-Cambium payload: Cambium and the Transgenic Arabidopsis Gene Expression System (TAGES). TAGES will use Arabidopsis thaliana with sensor promoterreporter gene constructs that render the plants as biomonitors of their environment using real-time nondestructive Green Fluorescent Protein (GFP) and traditional postflight analyses. Cambium Run 1 seeks definitive evidence that gravity has a direct effect on the cambial cells which contribute to reaction wood formation in trees, e.g., willow (Salix babylonica). Cambium Run 2 is an investigation of tree response to microgravity and the adaptation of white spruce (Picea glauca) to the space environment. Both experiments are conducted in the NASA developed Advanced Biological Research System (ABRS) ISS facility.

POINTS OF CONTACT

CSA Program Manager **NASA Project Manager** Luc Lefebvre David R. Cox



Bob Thirsk Harvesting Cambium Plants

Jeff Williams Harvesting TAGES Plants

Hardware	 - ABRS: 1 MLE replacement middeck/EXPRESS facility. Uses mdk rear breathing and EXPRESS AAA, MTL, Ethernet, video, commanding. ABRS to remain on ISS. - Approximately 1 MLE of ambient stowage per ascent (include both experiment & support hardware, some items limited life). - Cold stow required for all ascents and descents. 					
Facility/Interfaces	Advanced Biologic	cal Researc	h System (ABRS)			
Late access	<l-28 hrs<="" th=""><th colspan="3">ABRS, and limited life stow items.</th></l-28>	ABRS, and limited life stow items.				
Pre-flight	N/A					
In-flight: # of sessions	On 3 flights: Runs 1, 2 & 3 38 days continuous operation for each run.					
Post-flight	N/A					
Early Retrieval	R+6 hrs	KFTs for Run 1, 2 & 3.				
Target Subjects	US and CSA astronauts as operators only.					
Total # of Subjects	Short-term	N/A				
Required	Long-term	N/A				
Total # of Subjects Collected so far	Short-term	N/A				
Collected so far	Long-term	N/A				
Ground reference	Yes	Asynchro	onous at KSC SLSL			

Advanced Plant Experiments on orbit-Cambium

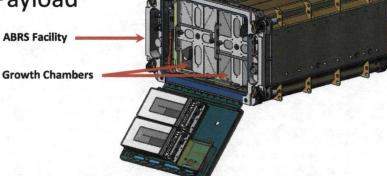
(APEX-Cambium) Payload













Ressources naturelles

PROPOSAL NUMBERS

CSA: ILSRA-04-122

NASA: 98-HEDS-02-299

SCIENCE TEAM

Rodney Savidge, Ph.D. Univ. of New Brunswick Robert Ferl, Ph.D.

Jean Beaulieu, Ph.D. Anna-Lisa Paul, Ph.D. Natural Resources Canada

Univ. of Florida

RESEARCH OBJECTIVES

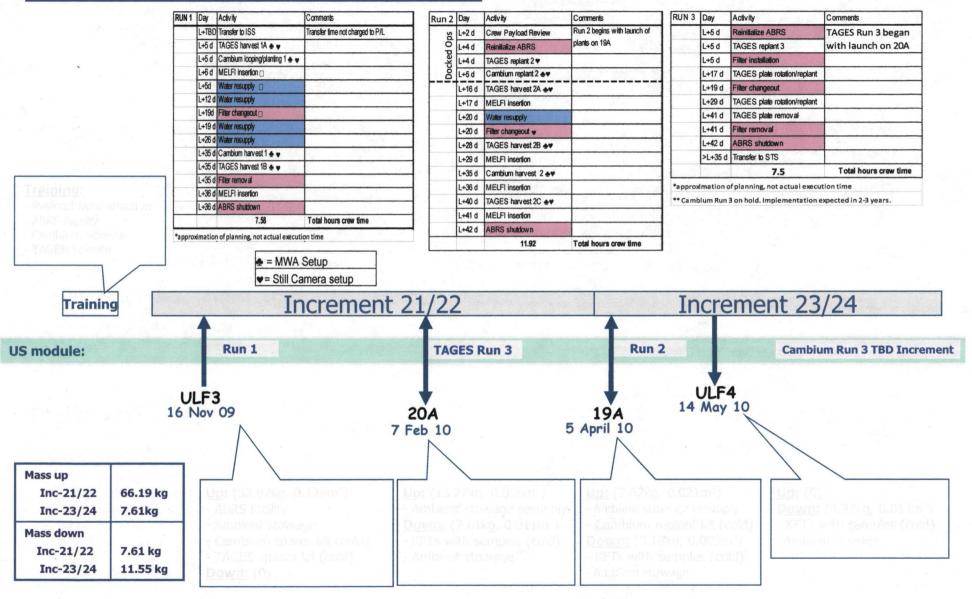
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Pre-flight	N/A					
In-flight: # of sessions	On 3 flights: Runs	On 3 flights: Runs 1, 2 & 3 38 days continuous operation for each run.				
Post-flight	N/A					
Early Retrieval	R+6 hrs	KFTs for	Run 1, 2 & 3.			
Target Subjects	US and CSA ast	ronauts as op	erators only.			
Total # of Subjects	Short-term	N/A				
Required	Long-term	N/A	N/A			
Total # of Subjects	Short-term	N/A				
Collected so far	Long-term	N/A				
Ground reference	Yes	Asynchr	onous at KSC SLSL			

APEX-Cambium - Increments 21/22, 23/24 scenario









- The initial TAGES 3A grow-out will use only a single Petri Plate installed into the imaged position (position 1) of the Green Fluorescent Imager as the primary objective of this experimental run is to obtain fluorescent images.
- Imager positions 2 6 will not be populated with Petri Plates at this time.





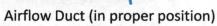
Airflow Duct Install



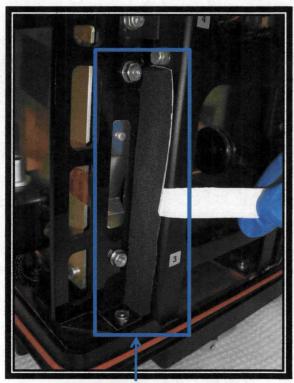


Kapton Tape covering seams

Kapton Tape covering gap (to seal off undesired airflow)



Green Fluorescent Imager Lens

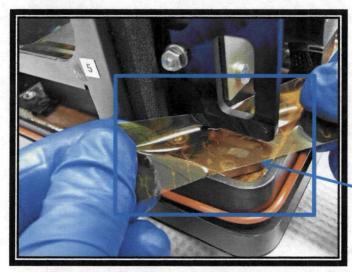


Foam Strip being installed on outer (lower) frame corners



Airflow Duct Install





Kapton Tape covering outside corners



Kapton Tape covering both outside corners, tucked in and not exposed from GFI frame



Kapton Tape covering outside slotted frame on the 3, 4 side



Stage 20A Operations



- Docked Operations:
 - ABRS Activation (only ERC 1)
 - ICL Flush (using improved procedure and kit H/W)
 - Airflow Duct installation into Green Florescent
 Protein (GFP) Imaging System (GIS) to prevent
 condensation on imaged surface of the Petri plate
 - Initiate Transgenic Arabidopsis Gene Expression
 System (TAGES) experiment Replant 3A in ERC 1



Stage 20A Operations



- Post-docked Operations after the conclusion of the TAGES 3A 12-day grow-out:
 - ABRS ICL Fuse Replacement with new fuse design
 - Potential ICL Pump Replacement as required
 - ICL Rear Flush using rear ICL QD (best possible flush)
 - Additional TAGES 3B and 3C 12-day grow-outs

NOTE: TAGES Run 3 science data is completely thru downlinked images from the ABRS GFP Imaging System and does not require harvested tissue; growth duration >12 days





Summary



- ABRS new EXPRESS sub-rack facility performing extremely well except for temperature control on ERC 2
- Thorough analysis performed and refurbishment plan developed in time for Stage 20A all while maintaining/maximizing the TAGES science return
- ABRS will be ready for CSA's next Cambium experiment on Stage 19A!

Advanced Plant Experiments on Orbit – Cambium (APEX-Cambium)

WBS: 825080.04.05.20.05



CSA PI: Dr. Rodney Savidge, University of New Brunswick U.S. PI: Drs. Robert Ferl & Anna-Lisa Paul. Univ. of Florida

PS: Dr. Howard G. Levine, NASA KSC

PM: David R. Cox, NASA KSC

Engineering Team: The Bionetics Corporation

Objective:

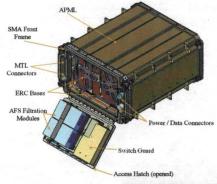
- Meet International agreements as an active member of the International Space Life Sciences Working Group (ISLSWG) and utilize U.S. flight hardware to support a Canadian Space Agency (CSA) investigation on reaction wood cell formation.
- Augment the CSA experiment with a U.S. investigator studying spaceflight stress effects on genetically modified plants using remote sensing and telemetric analysis techniques.

Relevance/Impact:

- APEX-CSA will maintain active U.S. participation in the ISLSWG as well as provide fundamental knowledge about reaction wood cell formation which will be extremely useful in the construction and paper pulp industries world wide.
- The U.S. investigation will demonstrate remote sensing biometric techniques for stress detection in living organisms during spaceflight. This knowledge will be extremely useful for future transit and surface missions to the Moon and Mars.

Development Approach:

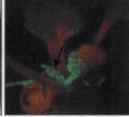
- Fly the NASA developed Advanced Biological Research System (ABRS) flight hardware on ISS. Perform Arabidopsis plant manipulations and harvests by the crew.
- Downlink images of the genetically modified Arabidopsis plants using the specialized Green Florescent Protein (GFP) imager integrated into ABRS.
- Preserve and return harvested tissue for ground analyses.





Clockwise: APEX hardware design elements, Looped Arabidopsis plant investigating reaction wood, GFP altered Arabidopsis plant images.





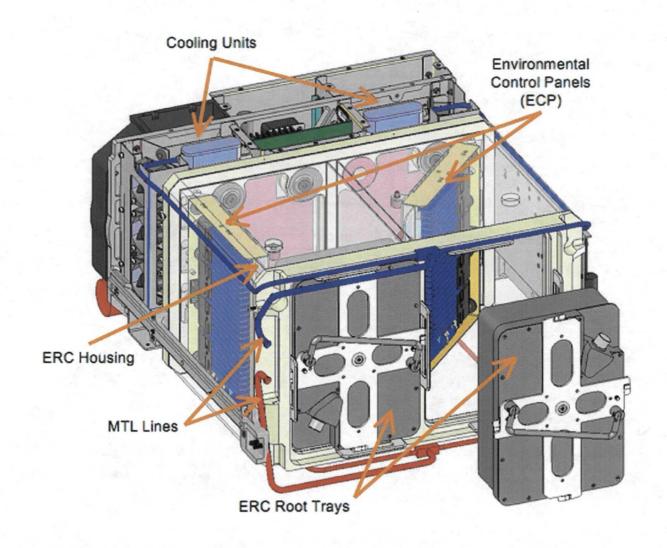
ISS Resource Requirements

Accommodation (carrier)	Middeck and EXPRESS Rk.		
Upmass (kg) (w/o packing factor)	50 (CSA Allocation)		
Volume (m³) (w/o packing factor)	2 MLE (CSA Allocation)		
Power (kw) (peak)	0.2 max (CSA Allocation)		
Crew Time (hrs) (installation/operations)	TBD (~15-30 CSA + ~5-10 NASA)		
Launch/Increment	1st Q FY09/Increments 18&19		

Project Life Cycle Schedule

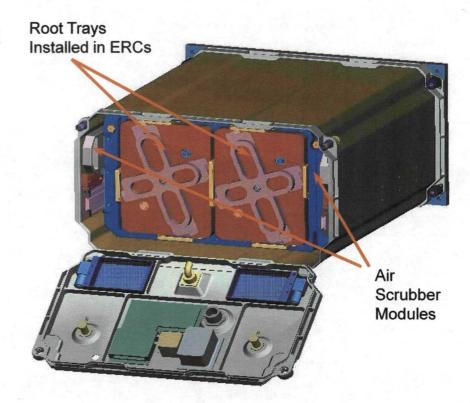
Milestones	SVT	Ph3 FSR	PVT	Launch	Ops	Return	Final Report
Actual/ Baseline	8/2007	10/2007	3/2008	1st Q FY09	ISS Incr-18/19	L+4-6 months	Return + 12m

ERC



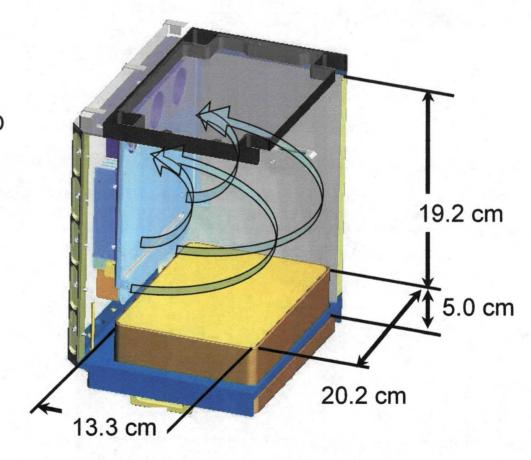
ABRS Overview

- Single middeck locker replacement facility plus stowage (filters, kits, etc.)
- Shuttle and EXPRESS compatible
- Easy specimen access and manipulation
- Can be used as a primary growth facility or up/down specimen transportation
- Two large, independent Environmental Research Chambers (ERCs)
- Condensate recovery and automated water delivery



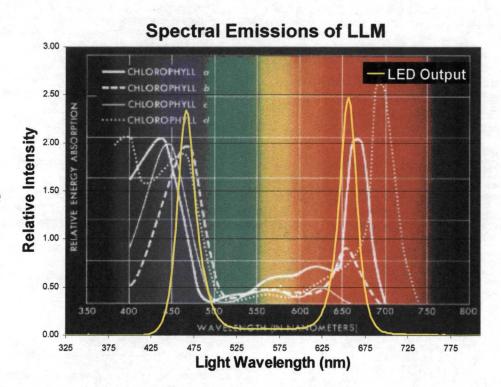
ABRS Environmental Research Chamber (ERC)

- Temperature: 23-26 °C (8 °C below ambient)
- PAR: 50-300 μmol/m²/sec
- Ethylene removal to below 25 ppb
- CO₂: controllable
- VOC removal
- RH: 60-90%
- Atmospheric humidity and root zone moisture control
- Volumetric air flow rate 6-15 cfm depending on RH
- Generic plant imaging
- GFP imaging capable



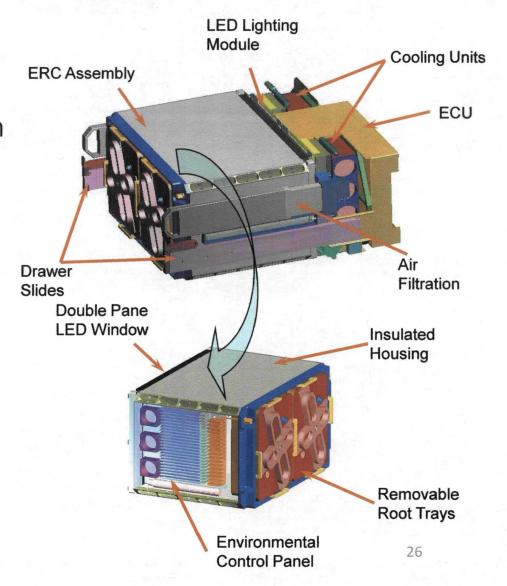
ABRS LED Light Module (LLM)

- LED growth lighting for improved efficiency
- Twin independent light banks
- Red, blue, green and white LEDs for full spectrum coverage
- Intensity of individual colors can be adjusted



ABRS Thermal Management

- Electronics Control Unit (ECU) located at the rear for heat rejection
- ERC assembly includes insulated sandwich-panel construction for chamber thermal isolation
- Growth chambers cooled by internal water loops-one loop per chamber
- Cooling unit TECs reject heat via rear-breathing (shuttle) and/or MTL (EXPRESS/ISS)



ABRS Contacts

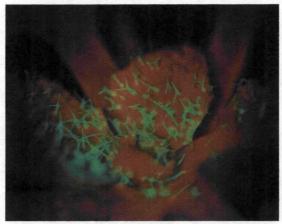
- ABRS Technical:
 Bill Wells
 Flight Experiments Manager
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 KSC, FL 32899
 321-861-3044
 Howard.W.Wells@nasa.gov
- NASA Project Management:

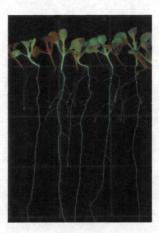
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 321-861-2980
 David.W.Reed@nasa.gov
- NASA Project Science:
 Howard Levine, Ph.D.
 ABRS Project Scientist
 Mail Code KT-B1
 KSC, FL 32899
 321-861-3502
 Howard.G.Levine@nasa.gov

TAGES (Transgenic Arabidopsis Gene Expression System)

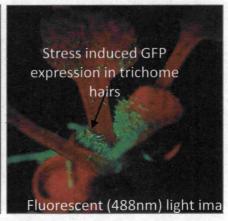
Principle Investigators: Dr. Robert J. Ferl and Dr. Anna-Lisa Paul University of Florida - Horticultural Sciences / Plant Molecular and Cellular Biology





Arabidopsis plant engineered with the Green Fluorescent Protein (GFP) gene reporters. The older plant at left is expressing Adh/GFP gene reporter in response to hypobaric (10 kPa) stress. This picture was featured on the cover of the Journal *Plant Physiology* (January, 2004) to illustrate our research article in that issue. The seedlings at right show plants expressing CaMV35s/GFP and Adh/GFP on the surface on an agar nutrient plate.





An Arabidopsis plant engineered with the Adh/GFP gene reporter. The left hand panel shows the plant photographed in white light and the right hand panel shows the same plant photographed in short-wave blue light to illuminate GFP expression. The actively growing apical meristem region of the plant and the trichome hairs are very sensitive to changes in the environment.

Acronyms

AAA	Avionics Air Assembly	KFT	KSC Fixation Tube
ABRS	Advanced Biological Research	LED	Light-Emitting Diode
	System	LLM	LED Lighting Module
APEX	Advanced Plant Experiments on Orbit	MTL	Medium Temperature Loop
CSA ECU	Canadian Space Agency Electronics Control Unit	NASA	National Aeronautics and Space Administration
EIA	EXPRESS Integration Agreement	OCA	Orbiter Communications Adapter
ERC	Environmental Research Chambers	PAR	Photosynthetically Active
EXPRESS	Expedite the Processing of		Radiation
	Experiments to Space Station	ppb	parts per billion
GFP	Green Fluorescent Protein	PVP	Payload Verification Plan
GUS	ß Glucuronidase	RH	Relative Humidity
IDD	Interface Definition Document	STS	Space Transportation System
ISLSWG	International Space Life Sciences Working	TEC	Thermo-electric Cooler
ISS	Group International Space Station	VOC	Volatile Organic Compounds